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An Assessment Of Sediments From The Upper Mississippi River

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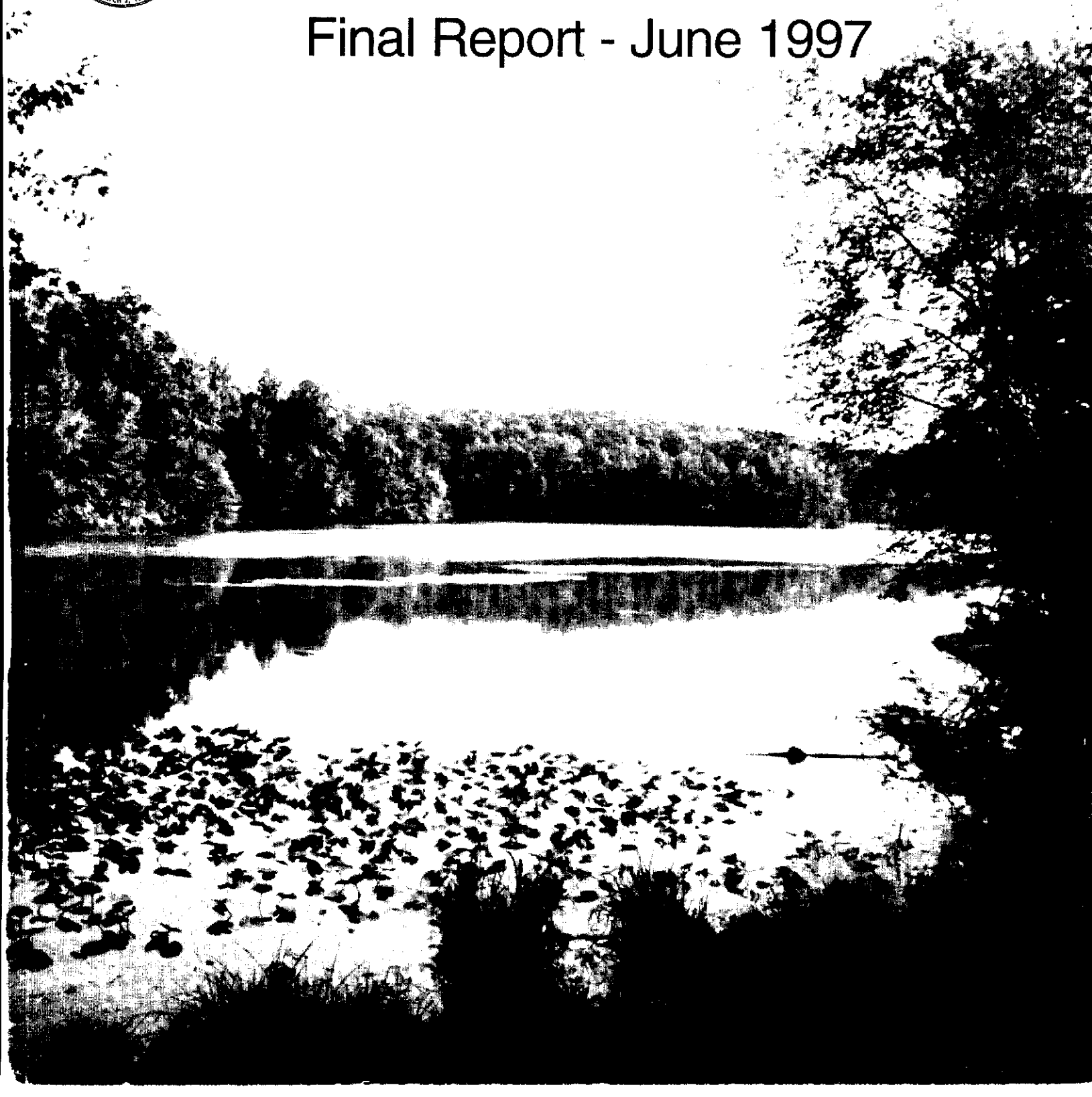

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ABSTRACT

The U.S. Geological Survey (USGS) has been monitoring the Upper Mississippi River (UMR) since 1987 to document the fate and transport of contaminats associated with sediments. The UMR is that part of the river upstream of the confluence with the Ohio River at Cairo, IL and consists of a series of 26 navigational pools created by a lock and dam system extending from Minneapolis, MN to St. Louis, MO. The navigational pools are shallow, lake-like areas which trap and store large quantities of fine-grained sediments during normal river flows. Concern with the redistribution of the river sediments arose after the flood of 1993. This project was designed to evaluate the current status of sediments in the UMR by: (1) measuring the concentrations of contaminants in sediments of the UMR, (2) evaluating the toxicity of sediments collected from the river, (3) determining the bioaccumulation of contaminants from UMR sediments using field-collected and laboratory exposed oligochaetes, and (4) determining the benthic community structure in fine-grain sediments within the river.

To conduct these assessments, sediment samples and benthic organisms were collected from 24 of the 26 navigational pools in the river and from one pool in the Saint Croix River. Two types of sediment samples were collected from the pools. One sediment sample was a composite of 15 to 20 sediment grabs along one to five transects across the downstream one-third of each pool (B samples). The other sediment sample was a composite of grabs from one station on one transect within each pool (C samples). The latter stations were selected based on historical chemistry data and the potential to collect oligochaetes. Samples were not collected from the main navigation channels. Chapter 1 of this report describes whole-sediment toxicity tests which were conducted for 28 days with the amphipod *Hyalella azteca*. Survival, growth and sexual maturation were the measurement endpoints. Toxicity tests were conducted with both the B and C sediment samples. Chapter 2 describes the bioaccumulation of contaminants from sediments using field-collected oligochaetes and 28-day bioaccumulation studies conducted in the laboratory with the oligochaete *Lumbriculus variegatus*. Bioaccumulation tests were conducted with 13 of the 24 C sediment samples. Chapter 3 assesses the benthic community in all 24 C samples. Using the Sediment Quality Triad approach, the status of UMR sediments was assessed by integrating sediment chemistry, laboratory toxicity tests and benthic community measurements.

In the toxicity tests, *Hyalella azteca* survival was significantly reduced in only one sediment sample (13B) relative to both a control and reference sediment. Growth of amphipods was also reduced in only one sediment sample (26C). Sexual maturation was not significantly reduced in any treatments. No correlations were observed between survival, growth or sexual maturation and any of the physical or chemical sediment characteristics. Using sediment chemistry and the Effect Range Median (ERM), 96% of the samples were classified as non-toxic (i.e. measured chemical concentrations rarely exceeded ERMs). Classifications using ERMs and sediment chemistry were consistent with the biological results from the *H. azteca* toxicity tests.

In the bioaccumulation tests, concentrations of contaminants were relatively low in native oligochaetes collected from the pools as well as in oligochaetes exposed to the sediments in the laboratory. Organochlorine pesticides were generally below detection in sediment and tissue samples. Only aliphatic and polycyclic aromatic hydrocarbons (PAHs) and total polychlorinated biphenyls were frequently measured above detection limits in oligochaete tissue and sediment

samples. Concentrations for a specific contaminant in laboratory-exposed and field-collected oligochaetes were similar within a station. About 90% of the paired PAH concentrations in laboratory-exposed and field-collected oligochaetes were within a factor of three of one another. With the detection limits used to analyze samples, contaminants were detected in tissue samples more often than in sediment samples. Concentrations of PAHs in oligochaetes collected from the pools or exposed in the laboratory to sediments from the UMR were up to 1000 times less than tissue concentrations measured in oligochaetes from highly-contaminated sites within the U.S. that our laboratory has previously studied.

The benthic community was dominated by oligochaetes and chironomids in 14 of the 23 sediment samples from the UMR and the one sediment sample from Saint Croix River. Fingernail clams comprised a large portion of the community in 3 of the samples and exceeded 1,000/m^2 in 5 of the samples. Total abundance values of invertebrates ranged from 250/m^2 (station 1C) to 22,389/m^2 (station 19C) and were comparable to previously reported values for the UMR. The frequency of chironomid mouthpart deformities was only 3% which is consistent with the incidence of mouthpart deformities from uncontaminated sediments. Correlations between benthic measures, sediment chemistry or other abiotic parameters exhibited few strong or significant correlations indicating benthic communities are most likely controlled by factors independent of contaminant concentrations.

The Sediment Quality Triad (Triad) is a weight-of-evidence approach used to assess the contamination of sediments by integrating sediment chemistry, laboratory toxicity testing and benthic community measures. Results from the Triad analysis indicated 88% of the samples were classified as not impacted based on sediment chemistry, laboratory toxicity and benthic measures. These results are consistent with the bioaccumulation study in which concentrations of contaminants in tissue were less than other U.S. sites that our laboratory has previously studied. In addition, pools in about the lower third of the river had lower sediment contaminant concentrations, less accumulation of contaminants in tissue, and greater taxa richness.

Sediments are often both a sink for water-borne contaminants and a source of contaminants to the overlying water. In addition, sediments may accumulate significant concentrations of contaminants even when water quality criteria are not exceeded. The results from the present study indicate that the UMR is not severely contaminated relative to other sites that have been studied in the U.S. Perturbations that may occur could be attributed to channelization, sedimentation from surface runoff or long term changes in the natural flow conditions of the river due to lock and dam construction. This study only conducted a partial assessment of the UMR sediments and included no assessment of river water. Further, this study was a one-time assessment that was conducted after a major flood event and does not evaluate temporal or spatial variability of sediment contamination within the pools. Future research on, or management of, the Upper Mississippi River should evaluate the limitations of this study.

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Introduction

The Mississippi River is the largest river system in the United States. Because of its location, the river receives contaminant inputs from a variety of industrialized and agricultural sources. The Upper Mississippi River (UMR), the stretch of river upstream from the confluence with the Ohio River at Cairo, IL, contains a series of 26 navigational pools created by a lock and dam system from St. Louis, MO to Minneapolis, MN (Rada *et al* 1990; Figure 1.1). These navigational pools are shallow lake-like areas which trap and store large quantities (1 to 4 cm/yr) of primarily fine-grained sediments during normal river flows (McHenry *et al* 1984; Nielsen *et al* 1984). Dredging activities, commercial navigation, recreational boating and natural resuspension processes can result in the remobilization of these sediments. Concern about the resuspension and transport of these sediments and the contaminants associated with them arose after the flood of 1993 (Moody and Meade 1995; Moody *et al* 1996).

The United States Geological Survey (USGS) has been monitoring the transport and degradation of pollutants in the UMR since the fall of 1987 (Moody and Meade 1995). Studies have monitored concentrations of contaminants in fish (Hora 1984; Wiener *et al* 1984), invertebrates (Beauvais *et al* 1995; Steingraeber and Wiener 1995), sediments (Bailey and Rada 1984; Wiener *et al* 1984; Rada *et al* 1990; Frazier *et al.* 1996; Ingersoll *et al* 1997) or a combination of the three (Peddicord *et al* 1980; Boyer 1984) in select pools in the UMR. However, little information was available on contaminant concentrations and toxicity in sediment samples throughout the entire pool system of the UMR.

Four studies were conducted to assess the nature and extent of sediment contamination in the navigational pools of the UMR: (1) contaminant concentrations were measured in sediments before and after the flood of 1993 (Moody *et al* 1996); (2) whole-sediment toxicity tests were conducted (this chapter); (3) whole-sediment bioaccumulation tests were conducted (i.e.; Chapter 2); and (4) benthic-community structure were evaluated (i.e.; Chapter 3). Sediment samples were collected from June 11th to July 5th, 1994 from pool 1 (near Minneapolis, MN) to pool 26 (near St. Louis, MO) of the UMR system (Figure 1.1). The objective of the study presented in this chapter was to assess the toxicity of sediments from navigational pools of the UMR system using 28-day toxicity tests with the amphipod *Hyalella azteca*, measuring for potential effects on survival, growth or sexual maturation.

Materials and Methods

Sample Collection, Handling, and Storage

Differential Global Positioning System (GPS) using a local reference was used to locate sampling stations in the upper pools (1-14) and the Saint Croix River. A differential GPS using the

navigational beacon near St. Louis, MO. was the reference to locate sampling stations in the lower pools (15-26). A 3.5 composite sediment sample was collected from each of the 26 navigational pools (pool samples designated as "B" samples; Moody *et al* 1996). These composite samples of surface (upper 10 cm) sediments were collected using a van Veen grab from 15 to 20 stations along one to five transects (typically 3 to 5 stations/transect) from the downstream one-third of each navigation pool (except pool 17) in the UMR and from one site in the Saint Croix River (SC) just upstream from its confluence with the Mississippi River in Wisconsin (Figure 1.1; Moody *et al* 1996). Samples were not collected from the main navigation channel which was assumed to contain coarser sediment that had been deposited for a short period of time. A 2-L subsample of the 3.5 L samples for toxicity testing and physical and chemical characterization were removed and placed in a 2-L high density polyethylene (HDPE) screw topped container. Samples were stored in a cooler at 4°C for 7 to 14 days on the research ship Acadiana, then shipped on ice to the Environmental and Contaminants Research Center (ECRC - formerly the Midwest Science Center) in Columbia, MO. Two 125-mL subsamples from each B sample were collected at the start of the toxicity tests for physical (grain size and TOC) and chemical (organic and metal) characterization.

A second composite sediment sample was also collected from each pool at one station on one of the transects (station samples designated as "C" samples). The individual stations (C samples) were selected based on historical chemistry data and the potential for the collection of large numbers of oligochaetes for bioaccumulation evaluations (Chapter 2). Station sediment samples (C samples) for toxicity and bioaccumulation (Chapter 2) testing were collected with a Ponar grab (529 cm^2 area). Each C sample was a composite sample collected from the upper 6 to 10 cm of the sediment surface within a 5-m radius area. A total of 35 to 80 L of sediment was collected from each C station. The sediment was then placed into a 120-L HDPE drum and homogenized on ship with a stainless steel auger on a hand-held power drill. Subsamples of these C samples were taken for (1) laboratory toxicity and laboratory bioaccumulation testing (10 L), (2) physical characterization (250 mL) and chemical characterization (250 mL for organics and 250 mL for metals) and (3) benthic invertebrate assessment (2 L). The remaining C sample was then sieved and native oligochaetes were collected for bioaccumulation analyses (Chapter 2). Sediment samples were stored in a cooler on the ship at 4°C for 7 to 14 days, then shipped on ice to the ECRC in Columbia, MO. Once at the ECRC, sediment samples were stored in the dark at 4°C until the start of the study. The control sediment (FLOR) used in the toxicity tests was a fine silt- and clay-particle size soil collected near St. Louis MO. This control sediment has been used in previous studies (Kemble *et al* 1994).

Culturing of Test Organisms

Amphipods were mass cultured at 23°C with a luminance of about 800 lux according to procedures outlined in Tomasovic *et al* (1995) using 80-L glass aquaria containing 50 L of ECRC well water (hardness 283 mg/L as $CaCO_3$, alkalinity 255 as $CaCO_3$, pH 7.8). Artificial substrates were also placed in the amphipod culture aquaria (six 20-cm diameter sections/aquarium of "coiled web material"; 3M Corp., Saint Paul, MN). Known-age amphipods were obtained by isolating mixed-aged adults in a 5-mm mesh (#35 US Standard size sieve) sieve in a pan

containing about 2 cm of well water. After 24 h, well water was sprinkled through the sieve, flushing <24-h-old amphipods into the pan below. These <24-h old amphipods were then placed into flow-through glass chambers for 10 d before the exposure began. Isolated amphipods were fed maple leaves and ground Tetramin® ad lib until the start of the test.

Toxicity Tests

Sediment Preparation: Sediment samples were re-homogenized in the laboratory using either a plastic spoon (for the B samples) or a hand-held power drill with a stainless steel auger (for the C samples). Subsamples were then collected for: (1) pore-water preparation, (2) physical and chemical characterizations, (3) toxicity testing, and (4) bioaccumulation testing © samples only; i.e., Chapter 2).

Water Quality: About 170 mL of pore water was isolated from each sample by centrifugation at 4°C for 15 min at 5200 rpm (7000 x G). A 50-mL subsample for total sulfide determination was removed from each sample and preserved with 0.1 mL of 2N zinc acetate solution (APHA 1985). Total dissolved sulfide was determined with an Orion EA940 Expandable ionAnalyzer, Orion 94-16 silver/sulfide electrode, and a Orion 90-02 double junction reference electrode. Dissolved oxygen (mg/L, with a YSI Model 54A oxygen meter and a YSI 5739 probe), temperature (°C) and conductivity (µs/cm @ 25°C with a Orion 140 S-C-T meter and a 014010 conductivity cell) were determined on the remaining volume. Subsamples of pore water were then removed for the following determinations: total ammonia (mg/L) with an Orion EA940, and Orion 95-12 ammonia electrode, alkalinity (mg/L, as $CaCO_3$) and pH with an Orion EA940 Expandable ionAnalyzer, Orion 917001 ATC probe, and Orion 8165BN combination pH probe, and total hardness as (mg/L, as $CaCO_3$) by EDTA titration. Unionized ammonia concentrations (mg/L, as NH_3) were calculated by adjusting total ammonia concentrations to pH and temperature using the formula presented in Thurston *et al* (1979). Hydrogen sulfide concentrations (mg/L) were calculated by adjusting the total dissolved sulfide concentrations to pH and temperature using the relationship presented in Broderius and Smith (1977).

Mean characteristics of porewater water quality (ranges in parentheses) are as follows: pH 7.45 (6.69 to 8.17); alkalinity 505 (244 to 852) mg/L; hardness 504 (148 to 852) mg/L; dissolved oxygen 5.04 (1.50 to 9.35) mg/L; conductivity 906 (380 to 1680) µs/cm @ 25°C; total ammonia 5.320 (1.210 to 22.700) mg/L; unionized ammonia 0.007 (0.000 to 0.025) mg/L; total sulfide 0.055 (0.000 to 0.569) mg/L; and hydrogen sulfide 0.023 (0.000 to 0.569) mg/L (Appendix 1.1).

The following parameters were measured in overlying test water on Day -1 (the day before amphipods were placed into the beakers) and at the end of each toxicity test: dissolved oxygen, temperature, conductivity, pH, alkalinity, total hardness, and total ammonia. Methods used to characterize overlying water quality in the whole-sediment tests were similar to the methods described for characterization of pore water. Dissolved oxygen, pH, and conductivity were also measured weekly. Temperature in the water baths holding the exposure beakers was measured daily. Overlying water pH, alkalinity, total hardness, conductivity and total ammonia measurements were similar among all stations, the control, and the in flowing test water (Appendix 1.2). Dissolved oxygen measurements were at or above acceptable levels (>40% of

saturation; ASTM 1995) in all treatments throughout the study (Appendix 1.2). Means (ranges in parentheses) of overlying water quality of each parameter are as follows: pH 8.07 (7.58 to 8.72); alkalinity 87 (59 to 151) mg/L; hardness 128 (111 to 160) mg/L; dissolved oxygen 6.70 (5.84 to 7.53) mg/L; conductivity 392 (359 to 428) µs/cm @25°C; total ammonia 0.416 (0.090 to 1.520) mg/L; and unionized ammonia 0.003 (0.000 to 0.012) mg/L (Appendix 1.2).

Toxicity Tests: All sediment tests were started within three months of sample collection from the field. Due to the number of samples collected, half of the samples (i.e., half of the sites) were randomly selected for the initial testing. The second set of sediment samples was tested after completion of testing of the first set of samples. Sediment samples for the toxicity tests were homogenized the day before animals were added to exposure beakers (Day -1), using procedures previously described.

Toxicity tests were conducted with *Hyalella azteca* for 28 days. Effects of exposure to sediments on survival, length, and sexual maturation of amphipods were measured (USEPA 1994; ASTM 1995). Each 300-mL beaker contained 100 mL of sediment and 150 mL of overlying water. The photoperiod was 16:8 h (light:dark) at a light intensity of about 500 lux. Four replicate beakers/sample were placed in a ventilated water bath maintained at 23°C. Each beaker received 1.0 volume additions/d of overlying water starting on Day -1 (Zumwalt *et al* 1994). The overlying water used in the sediment toxicity exposures was a reconstituted moderately hard water (hardness 95 mg/L as $CaCO_3$, alkalinity 65-70 mg/L as $CaCO_3$, pH 8.0-8.3; USEPA 1994). One diluter cycle delivered 50 mL of water to each beaker (diluters cycled every 8 h ± 15 min). Amphipods were acclimated to the test water over 6 h before exposures began by sequentially transferring animals at 2 h intervals into 50:50 and 25:75 mixtures of well water:test water, and then into 100% test water. Tests were started on Day 0 by placing 10 amphipods (10- to 11-d old) into each beaker. The water surface in each beaker was checked 15 min after organisms were placed in the beaker for floating organisms. Amphipods in each beaker were fed 3 mg of Purina Rabbit Pellets® in a water suspension three times a week for the first 7 days of the exposure, and 6 mg three times a week for the last 21 days of the exposure. If excessive mold (≥60% sediment surface) was observed on the sediment surface of any of the beakers in a treatment, feeding was withheld from all of the beakers for that treatment (the number of feedings withheld ranged from 0 to 5 depending on the treatment; USEPA 1994; ASTM 1996). Beakers were observed daily for the presence of animals, signs of animal activity (i.e., burrowing), and to monitor test conditions (i.e.; water clarity).

Amphipods were retrieved from each beaker at the end of exposures using procedures described in Kemble *et al* (1994). Surviving organism were combined into a scintillation vial and preserved in 8% sugar formalin for later measurement of length, and sexual maturation. A Zeiss® Interactive Digital Analysis System in combination with a Zeiss SV8 stereomicroscope at a magnification of 25x was used to measure amphipods following methods described in Kemble *et al* (1994). Amphipods were classified as either "mature male" or "not male" based on the presence of an enlarged second gnathopod (Kemble *et al* 1994). An enlarged second gnathopod of male amphipods was a consistent measure of sexual maturation (it is difficult to distinguish immature males from females at this age).

Chemical and Physical Characterization of Sediments

Acid-volatile Sulfides (AVS) and Simultaneously Extractable Metals (SEM): Subsamples of sediments were measured for acid-volatile sulfides (AVS) and simultaneously extractable metals (SEM) immediately after homogenization. Station samples (C samples) were collected on the boat and stored at 4°C until shipment to the laboratory. Pool samples (B samples) were collected in the laboratory immediately after sediment homogenization before the start of toxicity tests. Concentrations of AVS in sediment samples were determined using a silver/sulfide electrode following methods described in Brumbaugh *et al* (1994). Concentrations of SEM were determined using atomic spectroscopy following methods described in Brumbaugh *et al* (1994).

Percentage recoveries for inorganics from both blank and sediment extracts averaged 96%. The average range was from a low of 78% for antimony (spiked as sodium sulfide) in the sediment extract to a high of 110% for Zn in the sediment extract. The average duplicate coefficient of variation was 1.7% (6 compounds, n=2). Average duplicate coefficient of variation ranged from 0.2% for both Pb samples to 5.1% for S in one of the duplicate samples.

Organochlorine Pesticides (OCPs), Polychlorinated Biphenyls (PCBs), and Aliphatic and Polycyclic Aromatic Hydrocarbons (PAHs): Sediment samples (C samples) were prepared for the analyses of organochlorine pesticides (OCPs), polychlorinated biphenyls (PCBs), and aliphatic and polycyclic aromatic hydrocarbons (PAHs) by extracting twenty grams of sediment with acetone, followed by petroleum ether. A final acetone/petroleum ether extraction was done and the extracts combined, centrifuged and transferred to a separatory funnel containing sufficient water to facilitate partitioning of residues into petroleum ether portion. The petroleum ether was washed twice with water and concentrated by Kuderna-Danish to appropriate volume.

Organochlorine determination was conducted by transferring an aliquot of concentrated extract to a 1.6 g Florisil mini-column topped with 1.6 g sodium sulfate. Residues were eluted from the column in two elution fractions. The first fraction consisted of 12 mL of hexane followed by 12 mL of 1% methanol in hexane; the second fraction consisted of an additional 24 mL of 1% methanol in hexane. Quantification of residues in the two Florisil fractions and three silicic acid fractions was performed using a packed or megabore column and electron capture gas chromatography.

Hydrocarbon determination was conducted by transferring a second aliquot of the concentrated extract to a 20 g 1% deactivated silica gel column, topped with 5-g neutral alumina. Aliphatic and polynuclear aromatic hydrocarbon residues were fractioned by eluting aliphatics from the column with 100-mL petroleum ether (Fraction 1) followed by elution of aromatics using, 100-mL 40% methylene chloride/60% petroleum ether, followed by 50-mL methylene chloride (combined eluates, Fraction 2). Quantification of fraction 1 by capillary column, flame ionization gas chromatography was performed once the fraction was concentrated to appropriate volume. The silica gel (fraction 2) containing aromatic hydrocarbons was concentrated, reconstituted in methylene chloride and quantified by gas chromatography and mass spectrometry.

Average percent spike recovery for eighteen OCPs was 103% (n=2). The smallest average spike recovery was 68% for HCB while o,p'-DDE had the greatest average spike recovery (120%). Individual OCP concentrations were below minimum detection limits so duplicate

analyses were not evaluated. Average percent spike recovery for PAH compounds was 98% (29 compounds, n=2). Naphthalene (84%) had the smallest average percent recovery while fluoranthene had the greatest average spike recovery (110%). The average duplicate coefficient of variation was 12.6% (13 compounds, n=2). Average duplicate coefficient of variation ranged from 0% for multiple PAHs in both duplicate samples to 61% for benzo(a)pyrene in one of the samples.

Methods for the analyses of the B samples, detection limits and quality control are described in Moody *et al* (1996). Quality control of B sediment samples analyzed for PAHs included: (1) estimates of accuracy determined from the standard deviation of the percent recovery of deuterated compounds added to the extracts and calculated based on absolute area counts and external calibration, and (2) precision, based on the relative standard deviation of the absolute area of multiple analyses of a surrogate compound (Moody *et al* 1996). A list of all the PAHs and OCPs analyzed for in both sets of sediment samples (B and C) are listed in Appendix 1.3.

Physical Characterization of Sediments

Physical characterization of sediments included: (1) percentage water (Kemble *et al* 1993), (2) particle size using a hydrometer (Forth *et al* 1982; Gee and Bauder 1986; Kemble *et al* 1993), and (3) total organic carbon using a coulometric titration (Cahill *et al* 1987; Kemble *et al* 1993). All physical characterizations included analysis of duplicate samples. Differences in percentage water for duplicate samples ranged from 0% in treatments 2B, 7B, 13C, 14B and 18B to 7% in treatment 10C. Duplicate samples of control sediment, sucrose standards and blanks were analyzed when determining sediment total organic. Precision and accuracy of the coulometric technique used was tested against National Bureau of Standards and Standard Reference Materials (NBS-SRM) with an error of less than 0.03% of the excepted values (Cahill *et al* 1987). Differences between duplicates ranged from 0% in treatments 3B, 11B, 12B, 13C, 14C, 15C, 18C, 20C, 22C, 22B, 24C and 26C to 0.9% in treatments 5C, 9C and 26B.

Data Analysis and Statistics

Toxicity Tests: Before statistical analyses were performed, data for survival and maturation were arcsin transformed. Comparisons of mean survival and percentage sexual maturation were made using a one-way nested analysis of variance (ANOVA) with mean separation by Fisher's protected least significant difference test at alpha = 0.05 (Snedecor and Cochran 1982). Data for length had a normal distribution and were not transformed before statistical analysis. Comparison of mean body length was made using a one-way ANOVA with mean separation by Fisher's protected least significant difference test at alpha = 0.05 (Snedecor and Cochran 1982). A sample was designated as toxic when survival, growth, or sexual maturation were significantly reduced relative to the control and reference sediments. Sediments from pools 6 and 11 were chosen as reference sediment based on low concentrations of contaminants. Simple linear regression was used to compare physical and chemical sediment characteristics to amphipod survival, length or sexual maturation. All statistical analyses were performed with Statistical Analysis System (SAS) programs (SAS 1994).

Effects Range Median: Chemistry concentrations and toxicity endpoints were evaluated using 28-day *Hyalella azteca* Effect Range Medians (ERMs) reported by Ingersoll *et al* (1996) and Smith *et al* (1996). An ERM is defined as the concentration of a chemical in sediment above which effects are frequently or always observed or predicted for most species (Long *et al* 1995). The total number of individual ERMs exceeded with each sample was plotted against the sum ERM quotient (SERM-Q; where Q is equal to the concentration of each chemical in the sediment sample divided by the ERM for that chemical), similar to the toxic unit described by Canfield *et al* (1996), Ingersoll *et al* (1996) and Swartz *et al* (1997). We chose to evaluate sediment toxicity relative to nine ERMs which correctly classified >70% of the samples in Ingersoll *et al* (1996). These 9 individual ERMs tended to minimize Type I (false positive) and Type II (false negative) errors relative to other SECs reported by Ingersoll *et al* (1996). Due to insufficient chemistry data for chromium and total PCBs, only 7 of the 9 individual ERMs were used in this evaluation. These ERMs included: cadmium, lead, nickel, zinc, chrysene, benzo(a)pyrene, and benzo(g,h,i)perylene.

Results and Discussion

Toxicity Tests

Survival of amphipods was significantly reduced relative to the control and reference sediments only in the 13B treatment (Table 1.1). Body length of amphipods was significantly reduced relative to the control and reference sediments in only the 26C treatment (Table 1.1; Appendices 1.4 and 1.5). Sexual maturation was not significantly reduced in any treatments when compared to the control and reference sediments (Table 1.1; Appendices 1.6 and 1.7).

Indigenous organisms recovered at the end of amphipod exposures included oligochaetes, ostracods, clams, and a snail. Clam shells were present in many of the sediments; however, only a few live clams were retrieved at the end of the exposure. Pairs of amphipods were observed in amplexus in the control, 1-B, 2-B, 5-B, 6-C, 8-B, 8-C, 9-B, 10-B, 11-B, 14-C, 15-B, 18-C, 24-B, 24-C, and 26-B treatments, and gravid females were observed in the control, 11-B, 16-C, and 24-B treatments.

Although significant differences in survival of amphipods relative to the control and reference sediments were only observed in sample 13B, there was a relatively wide range in survival among the treatments. For example survival was below 70% in 13 of the 51 treatments (Table 1.1). Survival of amphipods in the control was acceptable (≥80%), however, survival in two of the four reference treatments (11C and 6B) was below 80%. Subsequent studies have found that the reconstituted water described in USEPA (1994) that was used to conduct this study does not consistently support adequate survival and growth of *Hyalella azteca* in 28-day exposures (McNulty 1995; Kemble *et al* 1996). Ingersoll *et al* (1997) retested sediment samples 4C, 11C, 14C, and 24C using well water as an overlying water and observed a mean survival of >90% in all of the samples with no substantial effects on growth, or reproduction of *H. azteca*. Survival of amphipods in these same sediments ranged from 48% to 63% in the present chapter (Table 1.1). Similarly, Benoit *et al* (1997) tested Station samples (7C, 9C, 13C, 22C, and 24C) in chronic toxicity tests with midge *Chironomus tentans* using a natural overlying water and did not observe

effects on survival, growth, emergence, or reproduction. Additional studies are ongoing to evaluate 28-day *Hyalella azteca* exposures using reconstituted waters.

Physical and Chemical Characteristics of Sediments

Physical and chemical characteristics of sediment samples are listed in Table 1.2. Sediment organic carbon content ranged from 0.2% for the sediment samples from Stations 6B and 20B to 5.2% for Station 10C. Organic carbon content in the control sediment was 1.2%. Percentage solids ranged from 21% in the sediment sample from stations 4C and 10C to 84% for the sediment sample from Station 20B. Classification of the sediment samples for grain size varied from pool to pool (i.e., loam (11C), sandy-loam (8B), silty-clay-loam (25 C and 22C)) while the control sediment was a silty-clay-loam (Table 1.2). Acid volatile sulfide levels ranged from 0.005 µmoles/g in the 1C sample to 63.0 µmoles/g in the 10C sample (Table 1.2).

Concentrations of simultaneously extracted metals in sediment samples are listed in Appendix 1.8. Sediment from sample 4C had the highest concentrations of extractable SEM Cd, Cu, Ni, and Pb. Sample 12C had the highest concentration of SEM Zn (Appendix 1.8). The sum SEM/AVS molar ratio in the present study was typically less than 1 (except the two samples from pool 1). This indicates the concentration of divalent metals listed in Appendix 1.8 were probably not high enough to result in toxicity of the samples (DiToro *et al* 1990). Concentrations of SEM Cd, Cu, Ni and Pb were highest in sediment samples from treatment 4C (Appendix 1.8). However, concentrations of SEM Cu and Pb were still below the ERMs reported by Ingersoll *et al* (1996; Figures 1.2 and 1.3).

Significant positive correlations were observed between SEM metals vs. TOC (Cu > Zn > Cd>Pb>Ni), SEM metals vs. percentage clay (Zn>Ni>Pb>Cu>Cd) and between SEM metals vs. percentage silt (Ni>Cu>Pb>Zn>Cd) when tested by Spearman's rho coefficient of rank correlation (Table 1.3). The significant negative correlation with sand and the positive correlation with clay and silt indicates that metals were concentrated in the finer sediment particles.

Concentrations of organochlorine pesticides (OCPs) in sediment samples are listed in Appendix 1.9. Concentrations of OCPs were below detection limits (0.01 µg/g) in all of the C samples except the 2C and SCC samples which had detectable concentrations of DDE and DDD (Appendix 1.9). Amphipod survival in the 2C sediment sample was 75%. However, despite having concentrations which were similar for both chemicals, survival of amphipods in the SCC sample was 90%. This indicates that the levels of DDE and DDD detected in these samples was not the sole cause of the lower survival observed in the 2C sediment sample. Concentrations of OCPs in the B samples were at or below detection limits for 10 of the 15 individual pesticides evaluated (Appendix 1.9). Concentrations for all 5 OCPs detected in the B samples were ≤0.079 µg/g dry weight and were below calculated ERMs (Smith *et al* 1996; Appendix 1.9).

Concentrations of polycyclic aromatic hydrocarbons (PAHs) in sediment samples are listed in Appendix 1.10. The highest concentrations were observed at Pool 1 and were generally lower in the downstream pools. Concentrations of PAHs in river sediments exceeded the Method Lower Limit of Quantitation (MLLQ; 0.03 µg/g) in at least one sediment sample for every PAH evaluated (except for 1-methylnaphthalene; Appendix 1.10). Concentrations of 4 of the 11 PAHs measured exceed at least one calculated ERM (Ingersoll *et al* 1996; Figures 1.4 and 1.5).

Elevated PAH concentrations in sediment samples were associated with sediment collected from pools near Minneapolis, MN. Concentrations of PAHs below pool 4 were similar in the remaining pools. Concentrations of fluoranthene exceeded the calculated ERM (0.175 µg/g) in 9 of the sediment samples from the Upper Mississippi River. Amphipod survival in these samples was above 75% in all but one of the samples (sample 4C which had a survival of 63%; Table 1.1). This would indicate that concentrations of fluoranthene in these samples had little or no effect on amphipod survival.

Comparisons of Sediment Characteristics to Toxicity Responses

Relationships of physical or chemical characteristics of sediments to toxicity were evaluated using rank correlation (Table 1.4). No significant correlations were observed between survival, growth or maturation and the measured physical or chemical characteristics of the sediment samples (Table 1.4). Additionally, no significant correlation was observed between the toxicity endpoints and concentrations of PAHs or OCPs normalized to total organic carbon concentrations (Table 1.5). Sediments from Pool 1 had the highest percent sand (>88%), but amphipod length and maturation were not reduced with exposure to 1B or 1C sediments relative to the control and reference sediments (Table 1.1). Similarly, the control sediment had the highest percent silt and clay relative to the other samples. Ingersoll and Nelson (1990), Kemble *et al* (1994), and Ingersoll *et al* (1997) also reported sediment particle size did not affect the response of *Hyalella azteca* in 28-d sediment exposures.

None of the 49 sediment samples exceeded any of the 7 individual ERMs. Use of these 7 ERMs correctly classified 47 of the 49 (96%) sediment samples from the UMR as non-toxic. The two samples incorrectly classified were both type II errors (false negative; toxic sample that does not exceed an ERM). This again may indicate something other than contaminants or contaminants not measured were the cause of the relatively wide range in survival among the treatments.

Additional ERMs for individual chemicals listed in Ingersoll *et al* (1996) and Smith *et al* (1996) were also evaluated. About 20% of the sediment samples exceeded at least one of these ERMs. However, use of these additional ERMs to classify samples as toxic or non-toxic resulted in increased Type I error (false positive; non-toxic sample that exceeds an ERM). As was the case when using only the seven ERMs, chemical concentrations from the two samples classified as toxic did not exceed any of the additional ERMs.

The prediction of sediment toxicity was also evaluated using a toxic quotient approach. A toxic quotient was calculated for each sample by first dividing the concentration of individual chemicals by their respective ERM and then summing each of the individual values (Canfield *et al* 1996; Ingersoll *et al.* 1996). In the present study, quotients for the seven chemicals listed above were used to calculate a toxic quotient for each sample (Table 1.2). Figure 1.6 plots the relationship between the frequency of ERM exceedances and the sum of the ERM toxic quotient. In the present study, the ERM toxic quotient was ≤ 2.6 and individual ERMs were not exceeded indicating the sediment samples from the UMR were relatively non-contaminated compared to sediments from areas of known contamination in the United States (Kemble *et al* 1994; Ingersoll *et al* 1996). A toxic quotient approach was also used in Chapter 3 using a quadrant frequency

analysis to evaluate the benthic community of the pools in the UMR system.

Summary

Toxicity tests using amphipods identified only two of the 49 sediment samples from the Upper Mississippi River system as toxic (a significant reduction in survival, growth or sexual maturation compared to the control and reference sediments). However, there was a relatively wide range in survival among the treatments. The overlying water used in this test was the reconstituted water described in USEPA (1994), which McNulty (1995) and Kemble *et al* (1996) have reported does not consistently support adequate survival of *Hyalella azteca* in 28-d sediment exposures. Survival of amphipods and midge was >90% in subsequent studies with sediments from the present study when natural water was used as the overlying test water (Benoit *et al* 1997; Ingersoll *et al* 1997). This would indicate that the reconstituted test water was a significant factor in the wide range of survival observed in the present study.

Effect Range Medians (ERMs) were used to evaluate the toxicity of contaminants associated with field collected sediments. ERMs correctly classified 96% of the UMR sediment samples as non-toxic. The two samples incorrectly classified were type II errors (false negatives). Again this indicates that factors other than contaminants or unmeasured contaminants may have been responsible for the variation in amphipod survival that was observed.

Concentrations of contaminants in sediments from the UMR were typically 10 to 100 times less than concentrations of contaminants in sediments previously associated with toxicity (Kemble *et al* 1994; Ingersoll *et al* 1996; Figure 1.7). This would indicate that the sediment samples from the UMR were relatively non-contaminated compared to other areas of known contamination across the United States.

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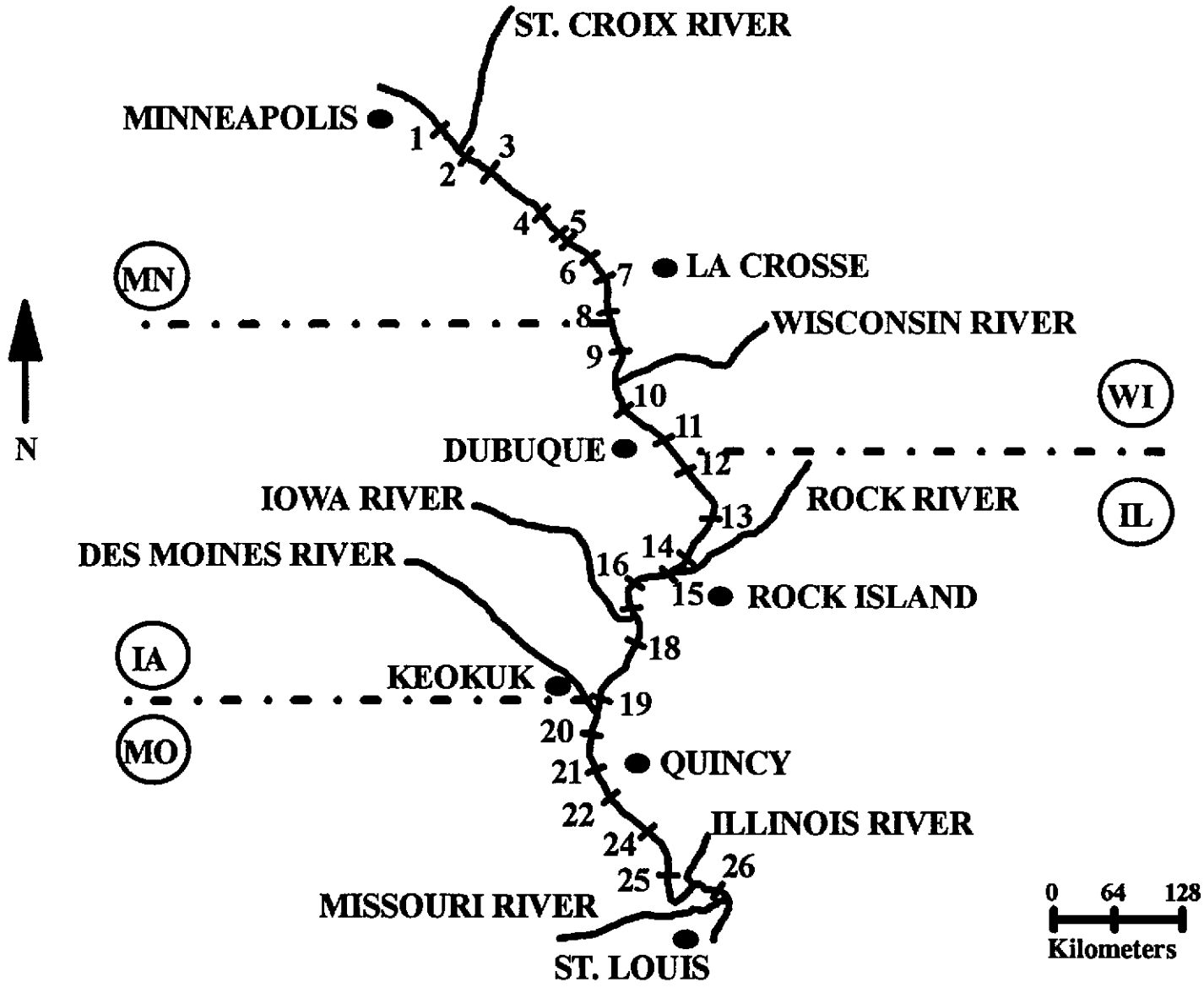

Fig. 1.1. Map of the Upper Mississippi River (UMR) from Minneapolis, MN to Saint Louis, MO.

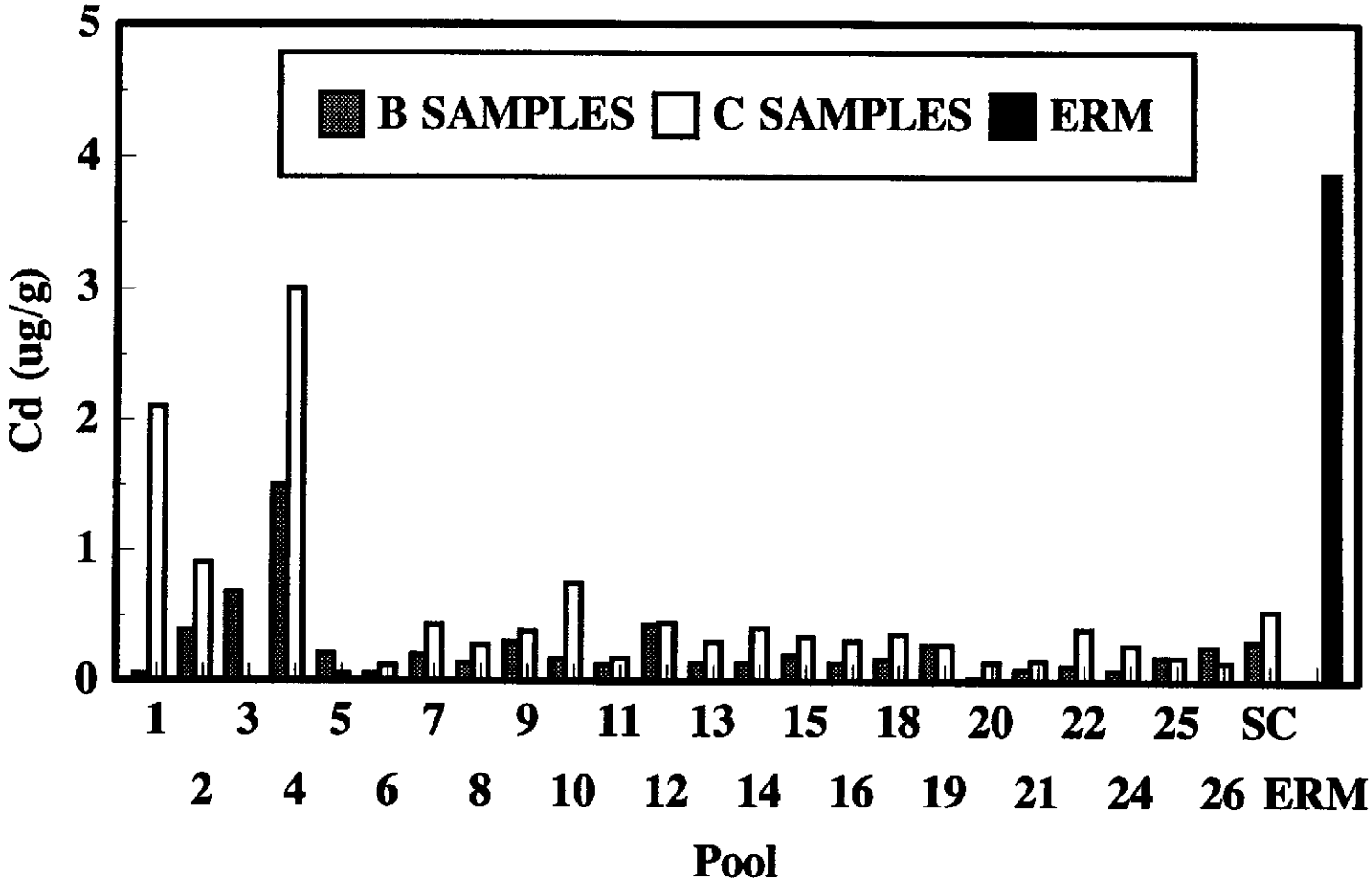

Fig. 1.2. Concentrations of Simultaneously Extracted Metal (SEM) Cd in UMR sediment samples compared to a Effect Range Median (ERM) for Cd.

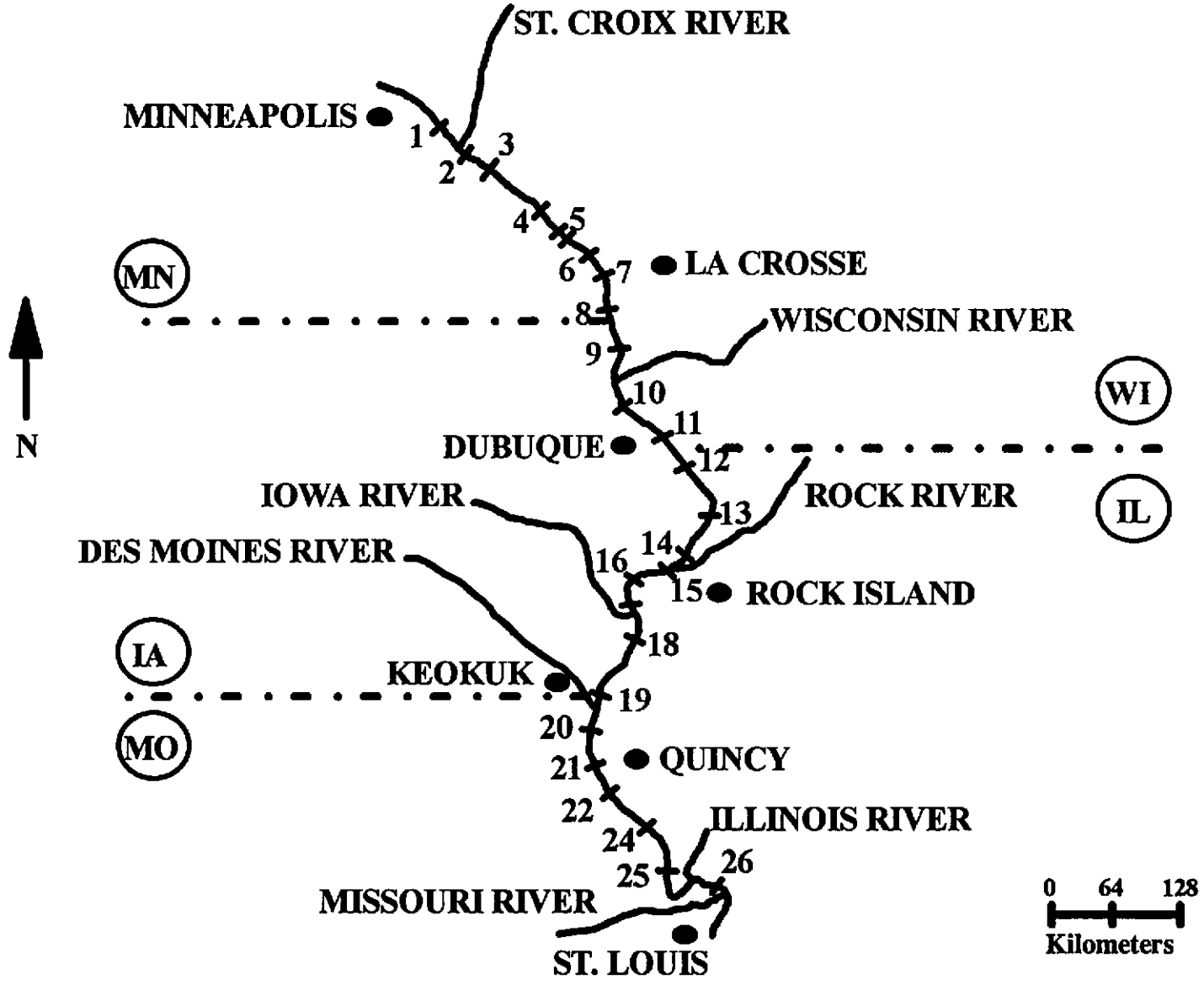

Fig. 1.1. Map of the Upper Mississippi River (UMR) from Minneapolis, MN to Saint Louis, MO.

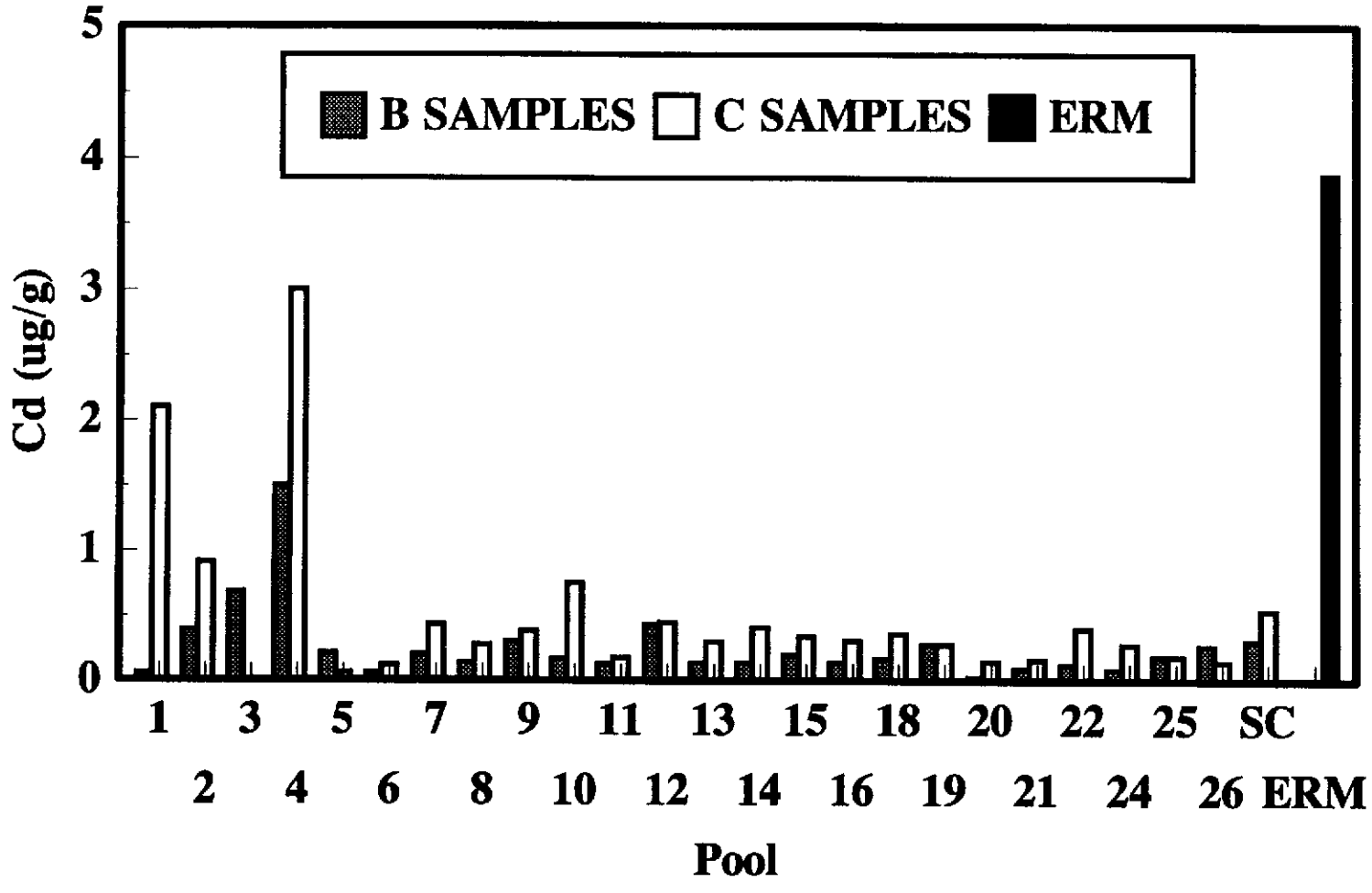

Fig. 1.2. Concentrations of Simultaneously Extracted Metal (SEM) Cd in UMR sediment samples compared to a Effect Range Median (ERM) for Cd.

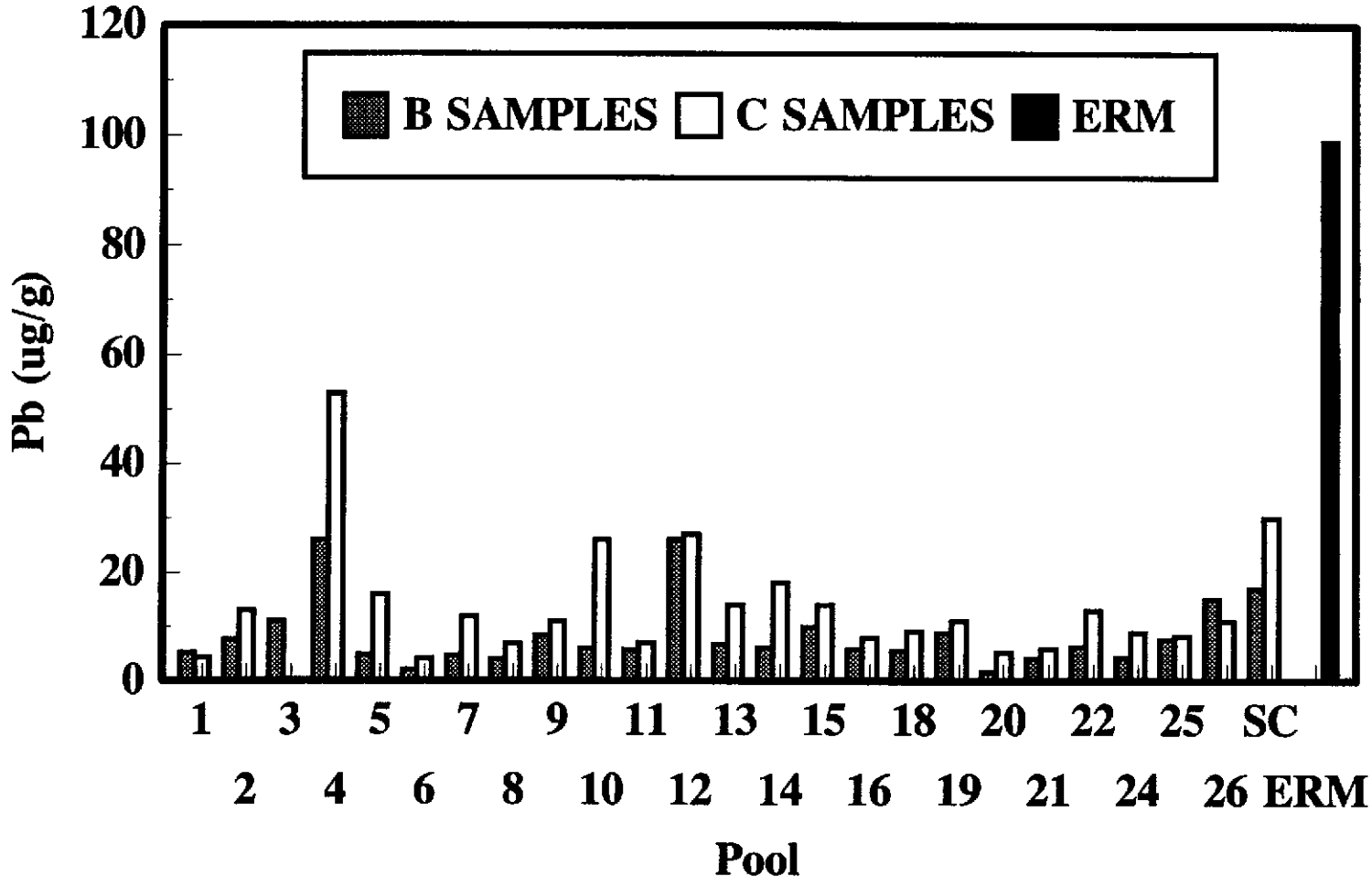

Fig. 1.3. Concentrations of SEM Pb in UMR sediment samples compared to a ERM for Pb.

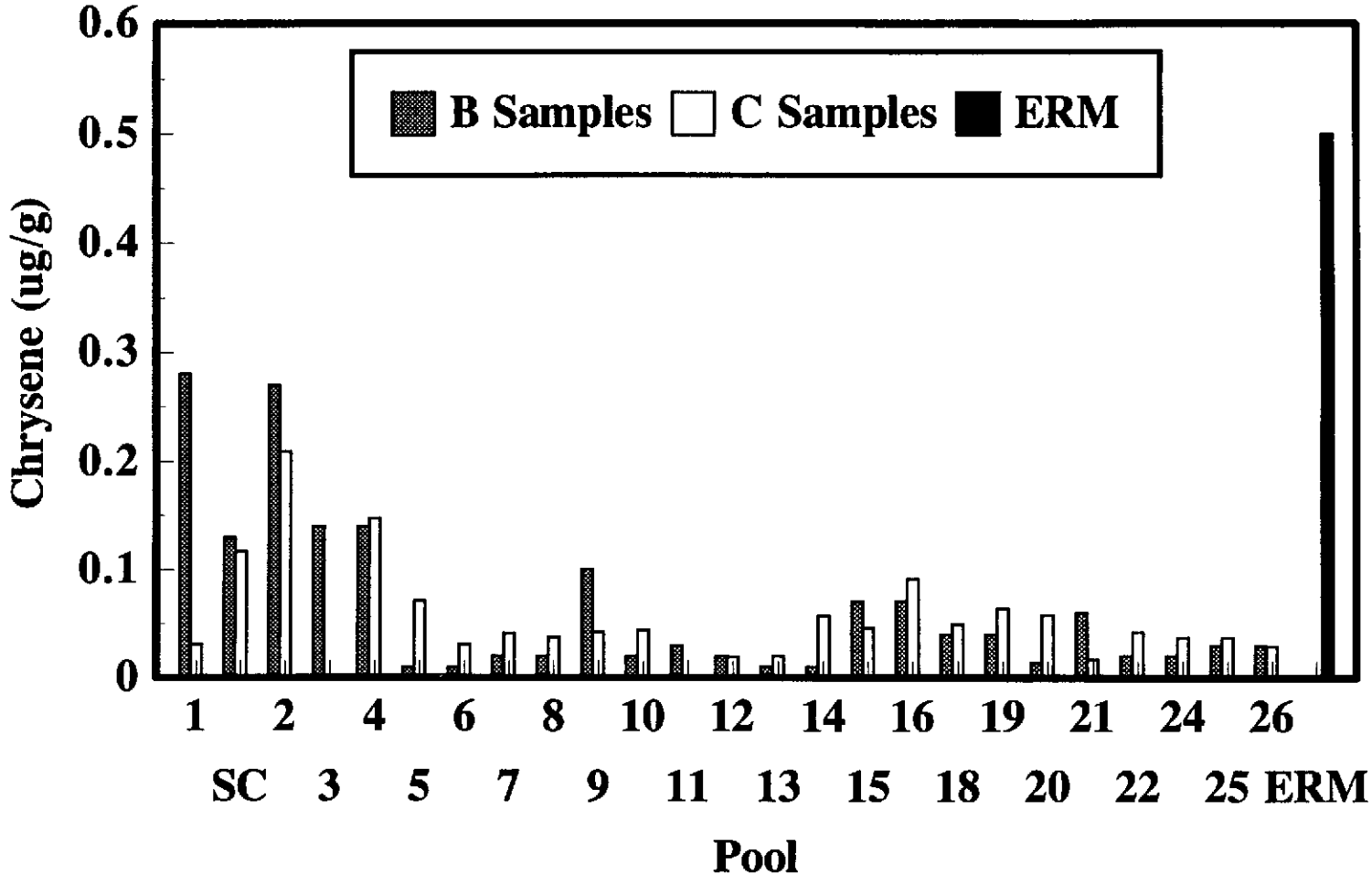

Fig. 1.4. Concentrations of Chrysene in UMR sediment samples compared to a ERM for Chrysene.

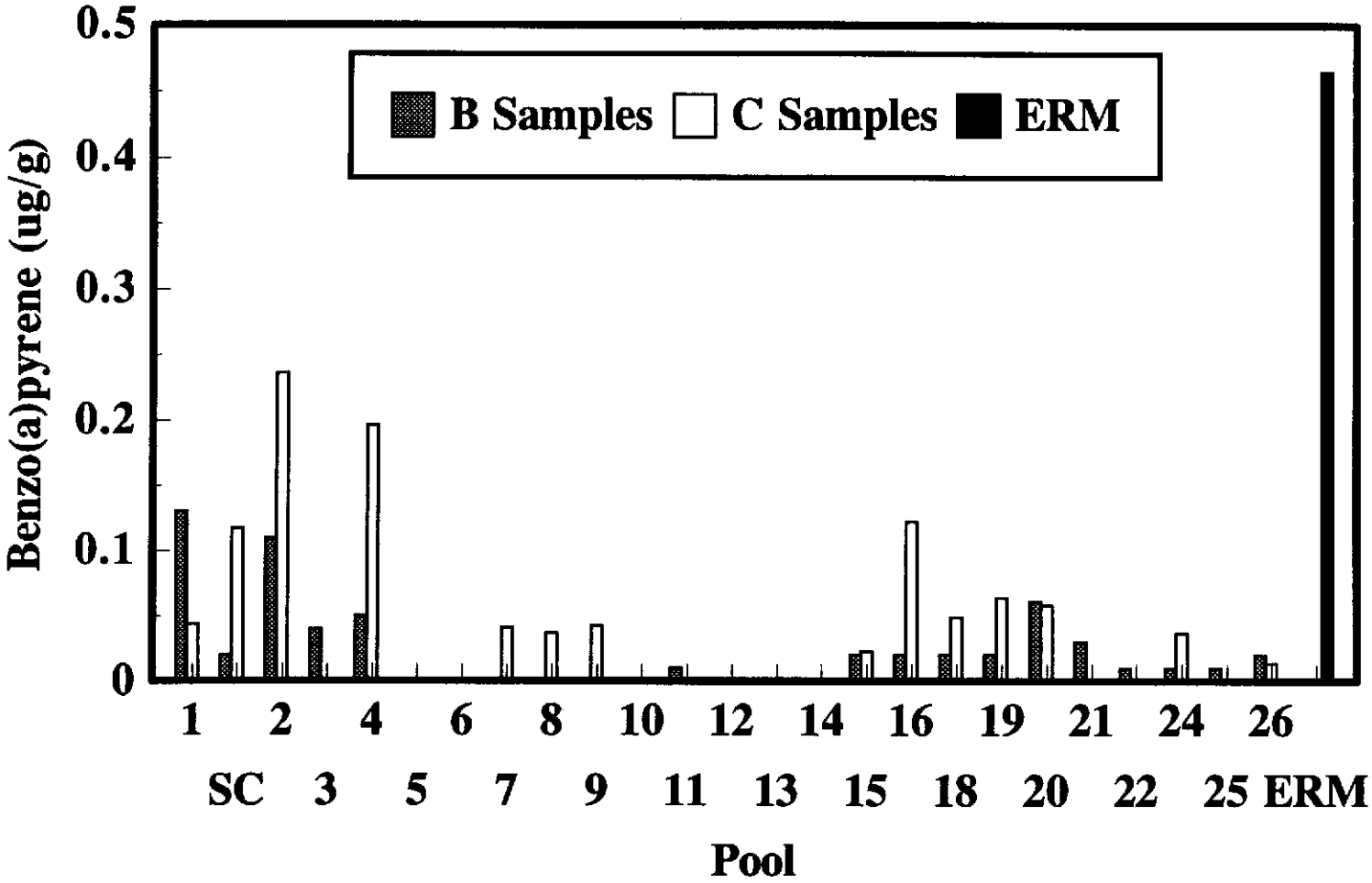

Fig. 1.5. Concentrations of Benzo(a)pyrene (BAP) in UMR sediment samples compared to a ERM for BAP.

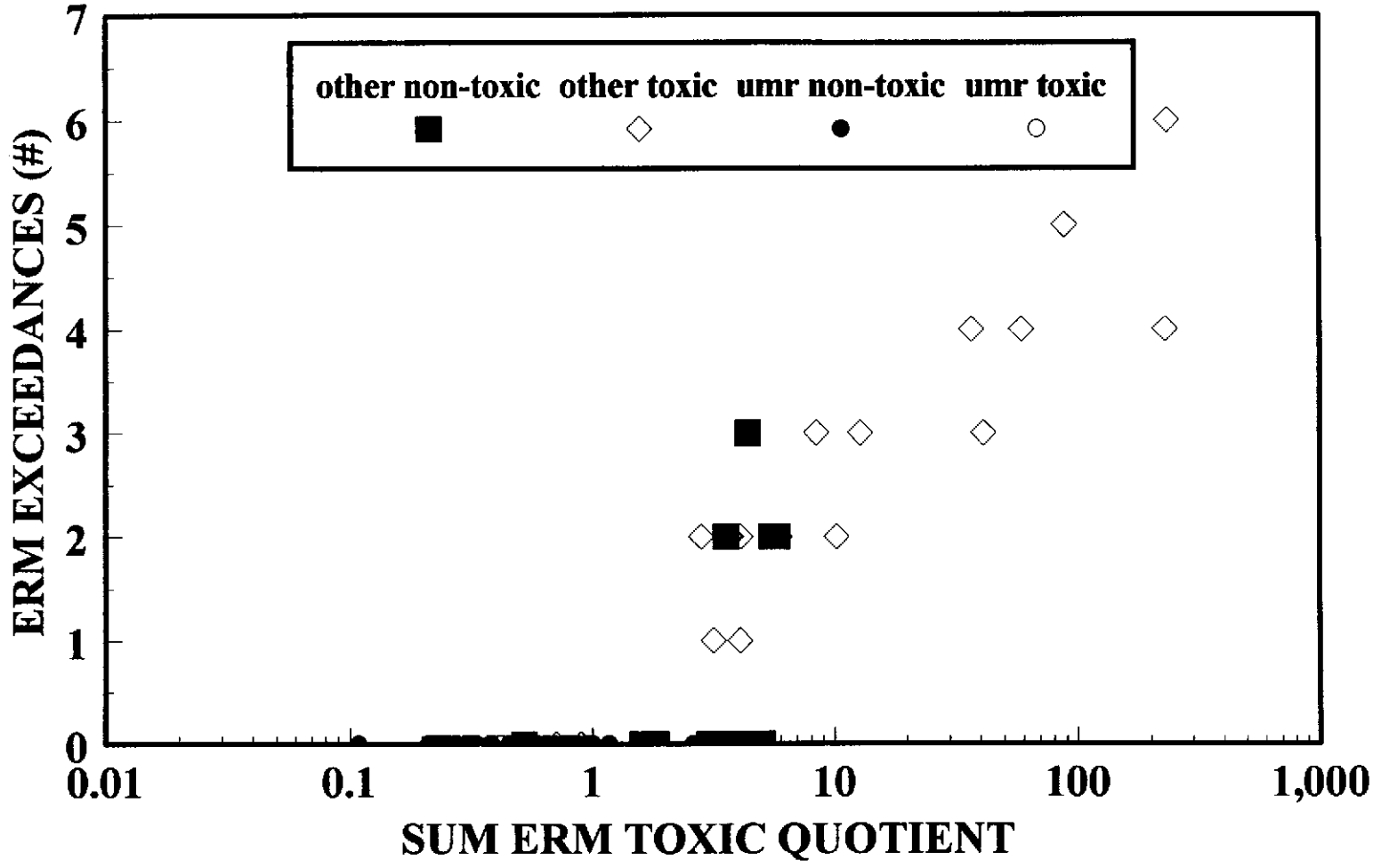

Fig. 1.6: Number of ERM exceedances for the 7 chemicals that correctly classified 70% of the samples compared to sum ERM toxic quotient.

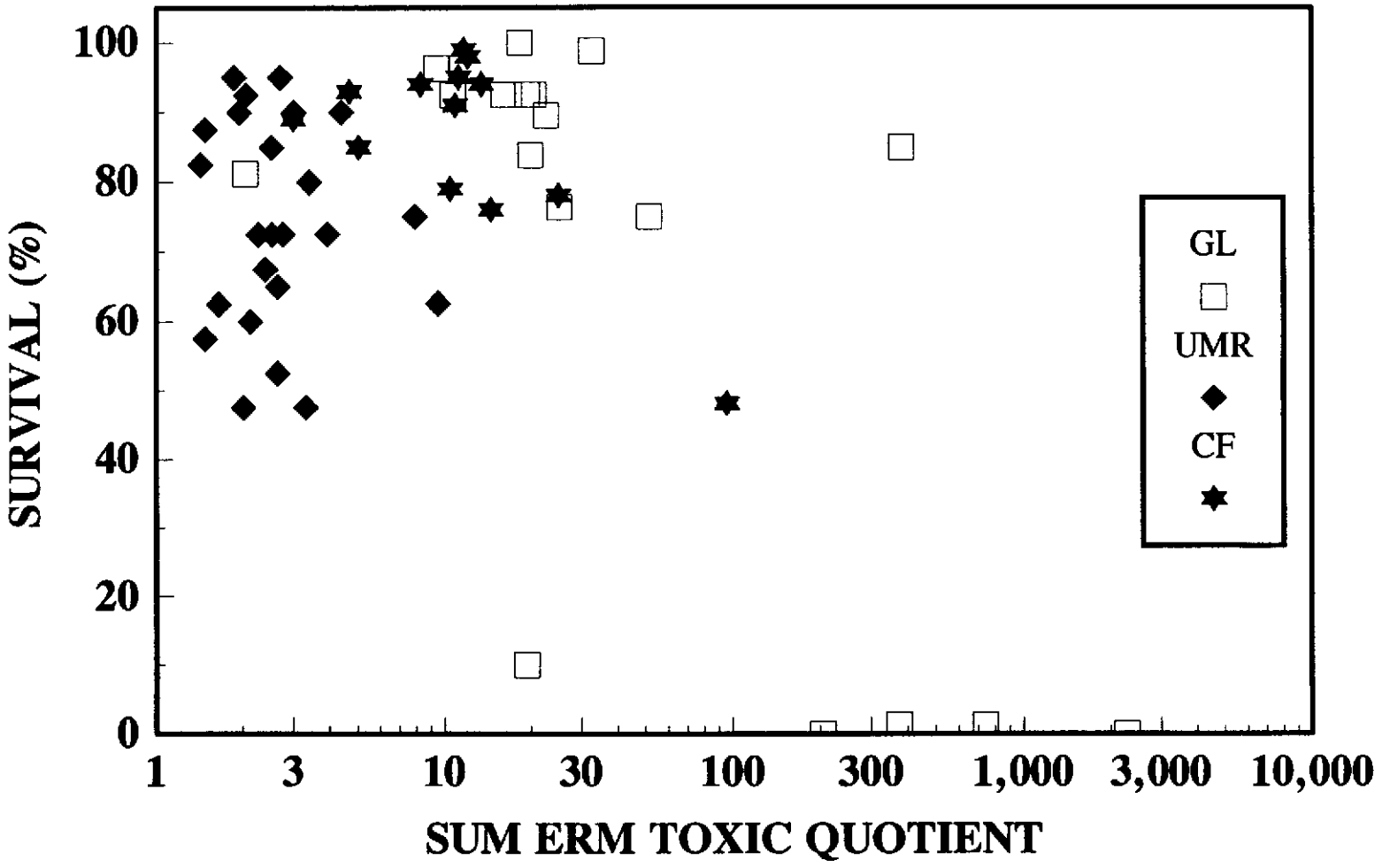

Fig. 1.7. Survival vs Sum ERM toxic quotients of sediment samples from the UMR compared to survival vs. sum ERM toxic quotients of sediment samples from the Great Lakes and the Clark Fork River and Milltown Reservoir MT.

Table 1.1. Results of the Upper Mississippi River sediment tests with *Hyalella azteca*. Means (Standard error of the means in parentheses) within a column and within a set of sample are significantly different ($p < 0.05$; n=4) from the control and reference sediment and are designated with an asterix.

Sample	Survival (%)	Length (mm)[1]	Mature Males (%)
1st set of samples			
Control	80.0 (4.08)	3.39 (0.16)	36.7 (8.91)
1B	92.5 (4.79)	3.66 (0.11)	39.1 (5.71)
1C	65.0 (5.00)	3.17 (0.11)	16.9 (6.90)
3B	95.0 (5.00)	4.27 (0.08)	44.9 (8.43)
5B	80.0 (7.07)	4.23 (0.06)	44.8 (10.30)
5C	80.0 (7.07)	4.06 (0.10)	21.6 (4.23)
8B	97.5 (2.50)	3.69 (0.09)	40.5 (7.72)
8C	92.5 (2.50)	4.09 (0.11)	32.3 (7.68)
10B	92.5 (7.50)	4.28 (0.09)	39.5 (18.49)
10C	72.5 (13.15)	3.86 (0.08)	34.4 (6.88)
11B (reference)	87.5 (2.50)	4.31 (0.07)	43.3 (11.57)
11C (reference)	57.5 (8.54)	3.61 (0.07)	32.8 (15.79)
12B	72.5 (9.46)	3.48 (0.07)	34.5 (3.00)
12C	85.0 (6.45)	3.78 (0.07)	32.4 (5.85)
15B	90.0 (4.08)	3.74 (0.08)	51.3 (11.46)
15C	72.5 (2.50)	3.59 (0.09)	34.0 (8.64)
16B	70.0 (9.13)	3.72 (0.08)	40.6 (6.56)
16C	90.0 (7.07)	3.83 (0.07)	30.0 (10.13)
21B	95.0 (2.89)	3.46 (0.06)	52.2 (6.08)
21C	87.5 (4.79)	3.87 (0.09)	51.4 (5.29)
25B	62.5 (13.15)	3.60 (0.11)	23.8 (10.51)
25C	62.5 (15.48)	3.63 (0.08)	29.6 (8.34)
26B	92.5 (4.79)	3.51 (0.09)	42.0 (6.82)
26C	90.0 (7.07)	2.88 (0.01) *	48.8 (11.30)
2nd Set of samples			
Control	97.5 (2.50)	2.59 (0.08)	5.9 (3.42)
2B	75.0 (8.66)	4.07 (0.11)	31.3 (6.25)
2C	75.0 (10.41)	3.47 (0.10)	43.8 (8.08)
4B	85.0 (6.45)	3.39 (0.10)	36.7 (13.72)
4C	62.5 (21.75)	3.35 (0.09)	12.1 (5.22)
6B (reference)	67.5 (17.02)	3.53 (0.09)	26.9 (9.21)
6C (reference)	82.5 (2.50)	4.08 (0.10)	54.5 (2.97)
7B	100.0 (0.00)	3.66 (0.06)	42.5 (10.31)
7C	95.0 (2.89)	3.70 (0.07)	35.5 (3.41)
9B	75.0 (10.41)	3.72 (0.09)	43.6 (6.47)
9C	67.5 (13.77)	3.65 (0.08)	32.8 (11.24)
13B	32.5 (7.50) *	3.87 (0.19)	18.8 (11.97)
13C	47.5 (10.31)	3.56 (0.11)	50.0 (9.64)
14B	65.0 (5.00)	3.85 (0.12)	31.6 (7.36)

Table 1.1. (continued)

Sample	Survival (%)	Length (mm)[1]	Mature Males (%)
14C	47.5 (7.50)	3.50 (0.12)	43.8 (15.72)
18B	77.5 (7.50)	3.57 (0.12)	50.0 (18.89)
18C	72.5 (17.97)	3.52 (0.09)	20.8 (7.50)
19B	85.0 (6.45)	3.31 (0.07)	40.2 (7.50)
19C	72.5 (7.50)	3.44 (0.07)	32.3 (15.91)
20B	82.5 (8.54)	3.43 (0.08)	11.9 (5.14)
20C	95.0 (2.89)	3.30 (0.06)	27.2 (10.74)
22B	85.0 (6.45)	3.79 (0.10)	24.4 (3.00)
22C	52.5 (10.31)	3.64 (0.11)	39.9 (14.20)
24B	87.5 (2.50)	3.61 (0.08)	34.4 (4.65)
24C	60.0 (8.16)	3.78 (0.12)	66.9 (14.19)
SCB	75.0 (10.41)	3.42 (0.10)	11.9 (7.89)
SCC	90.0 (4.08)	3.03 (0.06)	31.7 (5.60)

[1]starting body length of amphipods in the 1st set of samples was 1.05 mm (0.02 SE, n=11) and was 1.17 mm (0.04 SE, n=10) in the 2nd set of samples.

Table 1.2. Physical and chemical characteristics of sediments from the Upper Mississippi River at the start of whole-sediment tests. The sum ERM-quotient are also calculated for each sample.

Sample	Total Organic Carbon (%)	Solids (%)	Particle Size (%) sand	clay	silt	Sum ERM Quotient	Sediment Class
1B	0.3	76.5	88.6	9.3	2.1	1.17	Sand/Loamy Sand
1C	0.5	77.9	88.8	10.1	1.1	0.80	Sand/Loamy Sand
2B	3.6	61.3	53.5	25.5	21.0	1.66	Sandy Clay Loam
2C	3.3	45.0	15.4	43.1	41.5	1.58	Silty Clay
3B	2.7	53.2	27.5	23.5	49.0	1.00	Loam
4B	4.8	26.2	11.6	49.0	39.5	1.68	Clay
4C	5.0	20.8	33.4	39.8	26.9	2.60	Clay Loam
5B	1.6	61.5	53.6	19.4	26.4	0.22	Sandy Loam
5C	5.1	27.7	31.6	31.0	37.5	0.81	Clay Loam
6B	0.2	77.3	84.6	12.4	3.0	0.11	Loamy Sand
6C	0.7	70.2	78.1	13.6	8.3	0.28	Sandy Loam
7B	1.0	47.7	17.1	32.1	50.7	0.27	Silty Clay Loam
7C	2.3	62.1	56.5	16.8	26.7	0.65	Sandy Loam
8B	1.3	57.5	58.0	18.8	23.2	0.21	Sandy Loam
8C	2.2	55.5	11.5	37.0	51.5	0.47	Silty Clay Loam
9B	2.0	56.3	27.6	21.5	50.9	0.52	Silt Loam
9C	2.9	48.0	9.3	29.4	61.3	0.60	Silty Clay Loam
10B	1.2	55.2	59.6	36.9	3.5	0.25	Sandy Clay
10C	5.2	20.7	24.3	41.7	34.0	0.94	Clay
11B	1.3	59.8	46.1	18.8	35.1	0.28	Loam
11C	1.8	64.7	46.2	21.6	31.3	0.31	Loam
12B	2.0	54.2	20.0	20.9	59.1	0.77	Silt Loam
12C	2.3	54.9	15.3	21.4	63.3	0.84	Silt Loam
13B	1.8	65.4	33.2	23.1	43.7	0.24	Loam
13C	1.8	52.1	14.6	22.0	63.4	0.50	Silt Loam
14B	0.6	35.8	4.0	42.5	53.5	0.23	Silty Clay
14C	3.0	61.0	58.7	18.4	22.9	0.70	Sandy Loam
15B	1.4	46.9	0.0	23.0	77.0	0.48	Silt Loam
15C	1.9	59.0	41.5	20.5	38.0	0.59	Loam

Table 1.2. (continued).

Sample	Total Organic Carbon (%)	Solids (%)	Particle Size (%) sand	clay	silt	Sum ERM Quotient	Sediment Class
16B	1.2	67.0	53.7	18.9	27.4	0.39	Sandy Loam
16C	2.8	67.4	51.3	21.9	26.8	0.76	Sandy Clay Loam
18B	0.7	69.1	64.0	19.5	16.5	0.33	Sandy Loam
18C	1.7	62.6	21.8	23.8	54.5	0.56	Silt Loam
19B	1.9	54.9	33.8	29.4	36.9	0.47	Clay Loam
19C	2.3	49.2	7.6	34.0	58.4	0.66	Silty Clay Loam
20B	0.2	84.1	81.4	11.7	6.8	0.23	Loamy Sand
20C	0.8	73.5	52.1	22.0	26.0	0.45	Sandy Clay Loam
21B	0.5	69.9	64.0	23.5	12.5	0.32	Sandy Clay Loam
21C	1.1	59.0	44.4	25.8	29.8	0.30	Loam
22B	0.5	73.3	62.1	23.4	14.5	0.25	Sandy Clay Loam
22C	2.4	44.4	0.3	40.3	59.4	0.59	Silt Clay Loam
24B	0.7	74.6	57.5	23.0	19.5	0.21	Sandy Clay Loam
24C	1.7	57.1	30.7	22.0	47.4	0.50	Loam
25B	1.4	63.3	33.2	30.7	36.1	0.33	Clay Loam
25C	1.1	56.2	16.6	28.0	55.4	0.38	Silty Clay Loam
26B	2.0	54.5	24.1	33.5	42.5	0.51	Clay Loam
26C	0.7	72.6	43.5	27.0	29.5	0.42	Clay Loam
SCB	3.0	34.0	53.4	24.8	21.9	0.88	Sandy Clay Loam
SCC	4.3	26.6	36.1	25.5	38.5	1.17	Loam
FLORB	1.2	32.0	12.3	26.5	61.3	1.04	Silty Clay Loam
FLORC	1.2	32.0	12.3	26.5	61.3	1.04	Silty Clay Loam

Table 1.3. Spearman rank correlation for SEM Cd, Cu, Ni, Pb, and Zn with TOC, percent Sand, percent Silt, and percent Clay for Upper Mississippi River sediments (excluding the control sediment). All of the correlations listed below were significant ($p \leq 0.05$).

Element	TOC%	%Sand	%Silt	%Clay
Cd	0.826	-0.449	0.341	0.394
Cu	0.868	-0.556	0.563	0.468
Ni	0.808	-0.634	0.594	0.553
Pb	0.823	-0.583	0.434	0.549
Zn	0.854	-0.589	0.385	0.570

Table 1.4. Linear regression (r^2) of amphipod survival, length, or sexual maturation to sediment physical and chemical characteristics. None of the regression were significant ($p<0.05$).

	Survival	Length	Sexual Maturation
PW Total Ammonia	0.11	0.07	0.17
PW Unionized Ammonia	0.01	0.06	0.03
PW Total Sulfide	0.05	0.05	0.04
PW Hydrogen Sulfide	< 0.01	0.03	0.09
PW Alkalinity	< 0.01	0.09	0.08
PW Hardness	0.01	0.16	0.13
PW pH	0.02	< 0.01	< 0.01
PW DO	< 0.01	0.04	0.01
PW conductivity	0.01	0.13	0.03
AVS	0.11	< 0.01	0.02
Total Organic Carbon	0.02	< 0.01	0.02
Percent Sand	0.02	< 0.01	< 0.01
Percent Clay	0.01	< 0.01	0.01
Percent Silt	0.05	< 0.01	0.05
Percent Fines[1]	0.02	< 0.01	< 0.01
Percent Water	< 0.01	0.02	0.02
SEM Cd	0.03	0.04	0.11
SEM Cu	0.01	0.03	0.03
SEM Ni	< 0.01	< 0.01	< 0.01
SEM Pb	0.02	0.05	0.05
SEM Zn	0.02	0.01	0.02
Toxaphene	0.01	0.02	< 0.01
Mirex	0.05	0.04	< 0.01
DDD	< 0.01	0.10	< 0.01
DDT	0.05	0.04	< 0.01
DDE	< 0.01	0.12	0.00
Endrin	0.05	0.04	< 0.01
Dieldrin	0.05	0.04	< 0.01
Heptachlor epoxide	0.05	0.04	< 0.01
Lindane	0.05	0.04	< 0.01
Naphthalene	0.04	0.04	0.01
Acenaphthalene	0.04	< 0.01	< 0.01
Acenaphthene	< 0.00	0.02	0.07
Phenanthrene	0.02	0.01	0.01
Anthracene	< 0.01	< 0.01	< 0.01

Table 1.4. (Continued)

	Survival	Length	Sexual Maturation
Fluorene	0.01	0.01	< 0.01
Fluoranthene	0.04	< 0.01	< 0.01
Chrysene	0.02	< 0.01	< 0.01
Pyrene	0.03	< 0.01	< 0.01
Benzo(b)fluoranthene	0.04	< 0.01	< 0.01
Benzo(k)fluoranthene	0.01	0.07	0.03
Benzo(a)pyrene	0.01	0.05	0.04
Indeo(1,2,3,-cd)pyrene	< 0.01	< 0.01	0.01
Benzo(g,h,i,i)perylene	< 0.01	0.01	0.01

[1]Silt and Clay combined

Table 1.5. Linear regression (r^2) of amphipod survival, length, sexual maturation to sediment chemical characteristics normalized to organic carbon. None of the regressions were significant ($p<0.05$).

	Survival	Length	Sexual Maturation
Toxaphene	0.03	0.01	0.05
Mirex	0.02	0.04	< 0.01
DDD	0.01	0.10	0.01
DDT	0.01	0.02	0.01
DDE	< 0.01	0.03	< 0.01
Endrin	0.01	0.05	< 0.01
Dieldrin	< 0.01	0.07	< 0.01
Heptachlor epoxide	0.01	0.07	< 0.01
Lindane	0.02	0.05	< 0.01
Naphthalene	0.03	< 0.01	< 0.01
Acenaphthalene	< 0.01	< 0.01	< 0.01
Acenaphthene	0.02	0.02	0.02
Phenanthrene	0.01	< 0.01	< 0.01
Anthracene	< 0.01	0.06	< 0.01
Fluorene	< 0.01	< 0.01	0.01
Fluoranthene	0.04	< 0.01	< 0.01
Chrysene	0.14	0.07	< 0.01
Pyrene	0.05	0.06	< 0.01
Benzo(b)fluoranthene	0.06	0.02	< 0.01
Benzo(k)fluoranthene	0.01	0.03	< 0.01
Benzo(a)pyrene	0.04	0.07	< 0.01
Indeo(1,2,3,-cd)pyrene	< 0.01	0.05	0.01
Benzo(g,h,j,i)perylene	< 0.01	0.05	0.01

Appendix 1.1. Pore water quality for the whole-sediment tests with Upper Mississippi river samples.

Pool	pH	Alkalinity (mg/L)	Hardness (mg/L)	DO (mg/L)	Conductivity (µmho @25°C)	Total ammonia (mg/L)	unionized ammonia (mg/L)	Total Sulfide (mg/L)	Hydrogen Sulfide (mg/L)
01B	7.33	376	852	4.60	758	7.240	0.008	0.019	0.006
01C	7.69	292	280	7.60	541	1.540	0.004	0.000	0.000
02B	7.38	732	758	2.38	1354	5.900	0.007	0.011	0.003
02C	7.47	560	552	4.80	969	3.310	0.005	0.005	0.001
03B	7.50	624	664	6.60	1131	3.980	0.006	0.006	0.001
04B	7.59	ND	ND	4.23	779	2.745	0.005	0.302	0.056
04C	7.84	ND	ND	7.90	747	1.370	0.005	ND	0.000
05B	7.61	455	460	4.30	832	3.310	0.007	ND	0.000
05C	7.40	374	360	3.50	700	3.580	0.004	0.011	0.003
06B	7.82	432	404	6.20	830	3.470	0.011	0.099	0.012
06C	7.46	548	580	5.45	1031	11.400	0.016	0.040	0.009
07B	7.50	636	652	1.50	1186	5.640	0.009	0.000	0.000
07C	7.29	396	ND	4.35	765	6.150	0.006	0.009	0.003
08B	7.62	522	519	5.10	961	3.420	0.007	0.000	0.000
08C	7.45	596	600	5.60	1055	6.480	0.009	0.011	0.003
09B	7.43	835	750	6.15	1462	10.500	0.014	0.037	0.009
09C	7.41	ND	ND	4.00	1262	10.400	0.013	0.124	0.032
10B	7.48	488	480	4.05	912	4.730	0.007	0.002	0.000
10C	7.26	452	468	5.45	785	3.550	0.003	0.039	0.013
11B	7.50	ND	ND	5.60	930	4.120	0.006	0.000	0.000
11C	7.45	415	418	5.90	786	4.440	0.006	0.007	0.002
12B	7.40	568	620	6.00	1013	4.900	0.006	0.037	0.010
12C	7.20	710	600	4.60	1163	6.350	0.005	0.018	0.006
13B	7.24	852	808	4.60	1680	22.700	0.020	0.013	0.004
13C	7.41	ND	ND	4.75	897	8.070	0.010	0.011	0.003
14B	7.43	436	440	2.55	846	4.540	0.006	0.029	0.007
14C	7.53	ND	ND	4.35	636	4.290	0.007	0.465	0.096
15B	8.17	ND	ND	5.80	847	3.440	0.025	0.023	0.001
15C	7.47	364	360	7.20	671	2.360	0.003	0.012	0.003
16B	7.50	464	484	3.50	892	6.190	0.010	0.003	0.001
16C	7.40	ND	ND	5.40	998	6.970	0.009	0.031	0.008
18B	7.37	420	408	5.30	835	4.690	0.005	0.025	0.007

Appendix 1.1. Pore water quality for the whole-sediment tests with Upper Mississippi river samples (continued).

18C	7.44	340	348	2.95	652	3.180	0.004	0.007	0.002
19B	7.49	573	505	3.00	1027	5.440	0.008	ND	0.000
19C	7.33	ND	ND	3.90	1077	6.840	0.007	0.202	0.059
20B	0.00	ND	ND	ND	ND	2.750	0.000	0.569	0.569
20C	7.78	ND	ND	6.80	643	2.650	0.008	0.007	0.001
21B	7.45	ND	ND	5.30	1019	9.030	0.013	0.001	0.000
21C	7.31	ND	540	2.50	945	8.730	0.009	0.041	0.012
22B	7.41	495	475	5.10	934	6.370	0.008	0.000	0.000
22C	7.20	ND	ND	4.50	1109	12.300	0.010	0.159	0.057
24B	7.65	ND	ND	6.75	568	2.780	0.006	0.003	0.000
24C	7.45	ND	ND	6.40	708	ND	0.000	0.052	0.012
25B	7.34	ND	ND	1.50	869	3.260	0.004	ND	0.000
25C	7.42	440	352	5.60	768	2.270	0.003	0.008	0.002
26B	7.47	528	528	4.95	1003	5.520	0.008	0.007	0.002
26C	7.45	480	484	5.70	891	3.800	0.005	0.001	0.000
SCB	7.27	244	216	6.95	380	1.370	0.001	0.038	0.012
SCC	7.19	ND	148	6.50	386	1.210	0.001	0.057	0.021
FLOR	6.69	ND	ND	9.35	1176	1.410	0.000	0.001	0.000
Mean	7.45	505	504	5.04	906	5.320	0.007	0.055	0.023
Max.	8.17	852	852	9.35	1680	22.700	0.025	0.569	0.569
Min.	6.69	244	148	1.50	380	1.210	0.000	0.000	0.000
Std	0.21	141	159	1.60	246	3.649	0.005	0.114	0.084
Median	7.45	480	484	5.10	892	4.440	0.007	0.012	0.003

Appendix 1.2. Mean measured overlying water quality for the whole-sediment tests with Upper Mississippi river samples. Water quality was conducted on Days 0, 7,14,21, and 27.

Pool	pH	Alkalinity (mg/L)	Hardness (mg/L)	DO (mg/L)	Conductivity (µmho @25°C)	Total ammonia (mg/L)	unionized ammonia (mg/L)
01B	7.77	78	115	7.20	375	0.303	0.001
01C	7.87	72	112	7.44	367	0.045	0.000
02B	8.14	102	139	6.32	419	0.634	0.004
02C	8.72	151	160	6.52	408	0.473	0.012
03B	8.16	84	128	6.50	402	0.190	0.001
04B	7.96	83	129	6.04	396	0.312	0.001
04C	8.09	76	125	7.04	386	0.171	0.001
05B	8.10	78	121	7.18	388	0.183	0.001
05C	8.36	79	122	7.20	363	0.308	0.004
06B	7.97	81	130	7.02	391	0.331	0.002
06C	8.01	122	124	6.05	396	0.981	0.005
07B	8.10	115	144	7.06	391	0.610	0.004
07C	8.00	80	126	6.80	378	0.684	0.003
08B	7.95	79	120	7.04	386	0.218	0.001
08C	8.04	88	127	6.36	400	0.453	0.002
09B	8.17	111	148	6.44	413	0.902	0.007
09C	8.08	93	148	6.40	402	0.857	0.005
10B	8.15	82	122	7.04	396	0.285	0.002
10C	8.01	83	124	6.86	392	0.268	0.001
11B	8.11	78	119	6.94	386	0.310	0.002
11C	8.16	75	120	7.02	398	0.228	0.002
12B	8.30	83	125	7.05	395	0.228	0.002
12C	8.28	84	125	6.74	394	0.292	0.003
13B	8.06	129	154	5.84	428	1.520	0.009
13C	8.09	101	135	6.24	398	0.808	0.005
14B	8.02	94	135	6.21	403	0.570	0.003
14C	8.09	87	136	6.24	403	0.397	0.002
15B	8.20	78	117	6.88	400	0.160	0.001
15C	7.92	80	122	6.50	385	0.157	0.001
16B	8.15	81	126	6.76	412	0.324	0.002
16C	8.18	85	127	6.72	399	0.350	0.003

Appendix 1.2. Mean measured overlying water quality for the whole-sediment tests with Upper Mississippi river samples (continued).

18B	7.97	86	129	6.00	399	0.480	0.002
18C	8.19	83	144	6.40	391	0.354	0.003
19B	8.10	98	140	6.26	411	0.842	0.005
19C	8.07	94	140	5.92	411	0.593	0.003
20B	8.08	79	127	7.06	388	0.090	0.001
20C	7.97	78	119	7.02	374	0.369	0.002
21B	8.14	85	113	7.04	387	0.346	0.002
21C	8.10	83	124	6.26	395	0.423	0.003
22B	8.13	104	141	6.37	404	1.214	0.008
22C	8.09	94	140	6.24	410	0.587	0.004
24B	8.15	87	133	7.08	381	0.194	0.001
24C	8.14	96	136	6.76	397	0.377	0.003
25B	8.04	74	114	7.02	388	0.129	0.001
25C	8.05	76	116	6.94	385	0.220	0.001
26B	8.06	78	120	6.50	394	0.511	0.003
26C	8.03	76	121	6.54	383	0.254	0.001
SCB	7.74	60	113	6.88	364	0.199	0.001
SCC	7.86	66	111	7.04	359	0.199	0.001
FLOR B	7.71	59	115	7.53	373	0.220	0.001
FLOR C	7.58	71	116	7.24	369	0.091	0.000
Mean	8.07	87	128	6.70	392	0.416	0.003
Max.	8.72	151	160	7.53	428	1.520	0.012
Min.	7.58	59	111	5.84	359	0.090	0.000
Std	0.17	16	11	0.41	14	0.294	0.002
Median	8.09	83	125	6.76	394	0.324	0.002

Appendix 1.3. List of polycyclic aromatic hydrocarbons (PAHs) and organochlorines (OCs) analyzed for in the sediment samples from the Upper Mississippi River.

Polycyclic aromatic hydrocarbons

1. Naphthalene
2. 1-methylnaphthalene
3. 2,6-dimethylnaphthalene
4. Acenaphthene
5. Fluorene
6. Phenathrene
7. 1,-methylphenanthrene
8. Pyrene
9. Chrysene
10. 1,2-Benzanthracene
11. Perylene
12. Indeno(1,2,3-cd)pyrene
13. Benzo(g,h,i)perylene
14. C1-fluorenes
15. C2-fluorenes
16. C3-fluorenes
17. C1-phenanthrenes
18. C2-phenanthrenes
19. C3-phenanthrenes
20. C4-phenanthrenes
21. C1-fluoranthenes+C1-pyrene
22. C2-dibenzothiophenes
23. 2-methylnaphthalene
24. Biphenyl
25. Acenaphthalene
26. 2,3,5-trimethylnaphthalene
27. Dibenzothiophene
28. Anthracene
29. Fluoranthene
30. Benzo(b)fluoranthene
31. Benzo(k)fluoranthene
32. Benzo(e)pyrene
33. Benzo(a)pyrene
34. 1,2,5,6-dibenzanthracene
35. C1-naphthalenes
36. C2-naphthalenes
37. C3-naphthalenes
38. C4-naphthalenes
39. C1-dibenzothiophenes
40. C3-dibenzothiophenes
41. C1-chrysenes
42. C2-chrysenes
43. C3-chrysenes
44. C4-chrysenes

Organochlorines

1. Lindane
2. Heptachlor
3. Aldrin
4. Heptachlor epoxide
5. Chlordane
6. Endo
7. Dieldrin
8. DDE
9. Endrin
10. Perthane
11. DDD
12. DDT
13. Methoxychlor
14. Mirex
15. Toxaphene
16. o.p' DDE
15. HCB
16. alpha BHC
17. beta BHC
18. delta BHC
19. Oxychlordane
20. gamma Chlordane
21. trans-nonachlor
22. PCB 1242
23. PCB 1248
24. PCB 1254
25. PCB 1260
26. alpha Chlordane
27. o.p' DDD
28. cis-nonchlor
29. o,p' DDT

Appendix 1.4. Amphipod length data for the 1st set of sediment samples. Replication (Rep), Animal (individual animal number), and length (mean length for individual animal; n=2 measurements).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
ARCH	1	NA	0.946	1C	2	A	3.610
ARCH	2	NA	1.077	1C	3	A	3.475
ARCH	3	NA	1.134	1C	4	A	3.783
ARCH	4	NA	1.092	1C	5	A	3.669
ARCH	5	NA	0.973	1C	1	B	2.794
ARCH	6	NA	1.024	1C	2	B	3.616
ARCH	7	NA	1.122	1C	3	B	3.102
ARCH	8	NA	1.086	1C	4	B	4.112
ARCH	9	NA	1.000	1C	5	B	3.078
ARCH	10	NA	1.051	1C	6	B	2.946
ARCH	11	NA	1.086	1C	1	C	2.157
1B	1	A	3.373	1C	2	C	2.656
1B	2	A	2.522	1C	3	C	2.943
1B	3	A	3.048	1C	4	C	2.271
1B	4	A	3.084	1C	5	C	3.445
1B	5	A	3.610	1C	6	C	2.695
1B	6	A	3.090	1C	7	C	2.531
1B	7	A	3.655	1C	1	D	2.725
1B	8	A	3.265	1C	2	D	3.433
1B	1	B	3.843	1C	3	D	4.076
1B	2	B	3.666	1C	4	D	3.616
1B	3	B	4.348	1C	5	D	3.454
1B	4	B	3.630	1C	6	D	2.656
1B	5	B	3.783	3B	1	A	4.595
1B	6	B	3.765	3B	2	A	4.456
1B	7	B	4.207	3B	3	A	4.441
1B	8	B	3.556	3B	4	A	3.690
1B	9	B	3.846	3B	1	B	5.497
1B	10	B	3.332	3B	2	B	4.119
1B	1	C	4.398	3B	3	B	4.885
1B	2	C	4.889	3B	4	B	4.985
1B	3	C	3.864	3B	5	B	4.388
1B	4	C	4.646	3B	6	B	4.077
1B	5	C	3.409	3B	7	B	4.607
1B	6	C	4.942	3B	8	B	4.030
1B	7	C	4.073	3B	1	C	4.296
1B	8	C	4.883	3B	2	C	4.118
1B	9	C	4.222	3B	3	C	4.335
1B	1	D	3.173	3B	4	C	4.036
1B	2	D	2.925	3B	5	C	4.935
1B	3	D	2.474	3B	6	C	4.068
1B	4	D	4.282	3B	7	C	4.027
1B	5	D	2.752	3B	8	C	3.879
1B	6	D	3.179	3B	9	C	3.891
1B	7	D	3.164	3B	10	C	3.891
1B	8	D	3.472	3B	11	C	3.923
1B	9	D	3.215	3B	1	D	4.089
1C	1	A	3.170	3B	2	D	4.476

Appendix 1.4. Amphipod length data for the 1st set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
3B	3	D	4.053	5C	5	B	3.388
3B	4	D	4.181	5C	6	B	3.099
3B	5	D	3.805	5C	1	C	3.822
3B	6	D	3.778	5C	2	C	3.825
3B	7	D	4.867	5C	3	C	3.762
3B	8	D	3.920	5C	4	C	3.750
5B	1	A	3.974	5C	5	C	4.046
5B	2	A	3.825	5C	6	C	3.944
5B	3	A	4.037	5C	7	C	3.995
5B	4	A	4.103	5C	8	C	4.700
5B	5	A	4.754	5C	9	C	3.553
5B	6	A	4.249	5C	1	D	4.667
5B	7	A	4.357	5C	2	D	4.617
5B	8	A	3.669	5C	3	D	4.569
5B	9	A	4.112	5C	4	D	4.327
5B	1	B	4.467	5C	5	D	5.295
5B	2	B	3.965	5C	6	D	4.019
5B	3	B	4.198	5C	7	D	4.431
5B	4	B	3.834	5C	8	D	3.801
5B	5	B	4.524	5C	9	D	4.482
5B	6	B	4.404	8B	1	A	4.443
5B	7	B	4.216	8B	2	A	3.867
5B	8	B	4.070	8B	3	A	3.887
5B	9	B	3.971	8B	4	A	3.517
5B	1	C	4.682	8B	5	A	3.858
5B	2	C	4.088	8B	6	A	4.094
5B	3	C	4.387	8B	7	A	4.046
5B	4	C	4.987	8B	8	A	5.017
5B	5	C	4.216	8B	9	A	4.826
5B	6	C	5.265	8B	10	A	5.098
5B	7	C	3.732	8B	1	B	4.159
5B	1	D	4.422	8B	2	B	3.777
5B	2	D	4.088	8B	3	B	3.622
5B	3	D	4.159	8B	4	B	3.834
5B	4	D	4.261	8B	5	B	4.270
5B	5	D	4.091	8B	6	B	3.669
5B	6	D	4.162	8B	7	B	3.580
5C	1	A	3.251	8B	8	B	3.490
5C	2	A	4.216	8B	9	B	3.242
5C	3	A	4.073	8B	10	B	2.253
5C	4	A	3.230	8B	1	C	3.054
5C	5	A	4.512	8B	2	C	3.654
5C	6	A	5.157	8B	3	C	3.389
5C	7	A	3.765	8B	4	C	3.816
5C	1	B	4.192	8B	5	C	3.455
5C	2	B	3.696	8B	6	C	3.066
5C	3	B	3.974	8B	7	C	3.081
5C	4	B	3.672	8B	8	C	3.837

Appendix 1.4. Amphipod length data for the 1st set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
8B	1	D	3.352	10B	5	A	4.694
8B	2	D	3.675	10B	6	A	4.288
8B	3	D	3.151	10B	7	A	3.702
8B	4	D	4.045	10B	8	A	4.733
8B	5	D	3.557	10B	9	A	4.403
8B	6	D	3.374	10B	10	A	3.995
8B	7	D	3.560	10B	1	B	4.440
8B	8	D	2.940	10B	2	B	3.738
8B	9	D	3.922	10B	3	B	3.741
8B	10	D	3.075	10B	4	B	4.415
8B	11	D	3.373	10B	5	B	4.781
8C	1	A	4.283	10B	6	B	3.675
8C	2	A	4.238	10B	7	B	4.161
8C	3	A	3.581	10B	8	B	4.252
8C	4	A	4.027	10B	9	B	5.477
8C	5	A	3.916	10B	1	C	4.025
8C	6	A	2.807	10B	2	C	4.724
8C	7	A	3.367	10B	3	C	3.922
8C	8	A	2.458	10B	4	C	3.665
8C	9	A	4.142	10B	5	C	3.687
8C	1	B	4.241	10B	6	C	4.636
8C	2	B	4.253	10B	7	C	3.696
8C	3	B	3.858	10B	1	D	4.412
8C	4	B	5.099	10B	2	D	6.042
8C	5	B	3.831	10B	3	D	4.155
8C	6	B	3.678	10B	4	D	3.892
8C	1	C	4.136	10B	5	D	4.781
8C	2	C	4.184	10B	6	D	4.512
8C	3	C	4.127	10B	7	D	3.959
8C	4	C	3.467	10B	8	D	4.276
8C	5	C	4.524	10B	9	D	4.739
8C	6	C	3.876	10B	10	D	4.001
8C	7	C	3.461	10C	1	A	3.641
8C	8	C	3.587	10C	2	A	3.829
8C	9	C	4.425	10C	3	A	4.573
8C	1	D	4.460	10C	4	A	4.052
8C	2	D	4.346	10C	5	A	3.989
8C	3	D	4.322	10C	6	A	3.396
8C	4	D	3.654	10C	1	B	3.321
8C	5	D	4.747	10C	2	B	3.944
8C	6	D	4.383	10C	3	B	4.086
8C	7	D	5.024	10C	4	B	3.411
8C	8	D	5.575	10C	5	B	3.647
8C	9	D	5.015	10C	6	B	3.844
10B	1	A	4.594	10C	1	C	3.826
10B	2	A	4.104	10C	2	C	3.575
10B	3	A	3.959	10C	3	C	4.122
10B	4	A	3.632	10C	4	C	4.815

Appendix 1.4. Amphipod length data for the 1st set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
10C	5	C	4.691	11C	1	A	3.813
10C	6	C	3.947	11C	2	A	3.095
10C	7	C	3.620	11C	3	A	3.628
10C	8	C	3.638	11C	4	A	3.357
10C	9	C	4.270	11C	5	A	3.741
10C	1	D	3.807	11C	6	A	3.143
10C	2	D	4.255	11C	7	A	3.732
10C	3	D	3.650	11C	1	B	3.741
10C	4	D	3.811	11C	2	B	4.179
10C	5	D	2.858	11C	3	B	3.325
10C	6	D	3.623	11C	4	B	4.107
10C	7	D	4.180	11C	5	B	3.497
10C	8	D	3.547	11C	1	C	3.280
11B	1	A	4.589	11C	2	C	3.664
11B	2	A	4.051	11C	3	C	3.571
11B	3	A	4.333	11C	4	C	4.146
11B	4	A	4.164	11C	5	C	4.122
11B	5	A	4.152	11C	6	C	3.652
11B	6	A	4.262	11C	1	D	3.315
11B	7	A	4.066	11C	2	D	3.717
11B	8	A	3.950	11C	3	D	3.688
11B	1	B	3.828	11C	4	D	3.574
11B	2	B	4.220	11C	5	D	3.057
11B	3	B	5.000	12B	1	A	4.003
11B	4	B	3.768	12B	2	A	4.275
11B	5	B	4.119	12B	3	A	4.095
11B	6	B	4.244	12B	4	A	3.586
11B	7	B	3.408	12B	5	A	3.414
11B	8	B	3.661	12B	6	A	3.837
11B	9	B	4.235	12B	7	A	2.515
11B	1	C	4.357	12B	8	A	3.154
11B	2	C	4.057	12B	9	A	3.870
11B	3	C	4.878	12B	1	B	3.210
11B	4	C	4.351	12B	2	B	2.669
11B	5	C	5.122	12B	3	B	3.447
11B	6	C	4.408	12B	4	B	3.678
11B	7	C	4.351	12B	5	B	2.964
11B	8	C	5.006	12B	6	B	3.039
11B	9	C	5.116	12B	7	B	3.873
11B	1	D	3.479	12B	8	B	3.769
11B	2	D	4.116	12B	9	B	3.876
11B	3	D	4.661	12B	1	C	3.453
11B	4	D	4.432	12B	2	C	3.755
11B	5	D	4.577	12B	3	C	3.293
11B	6	D	4.360	12B	4	C	3.036
11B	7	D	5.027	12B	5	C	3.494
11B	8	D	4.137	12B	6	C	3.335
11B	9	D	4.217	12B	7	C	3.512

Appendix 1.4. Amphipod length data for the 1st set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
12B	1	D	3.663	15B	5	A	3.293
12B	2	D	3.352	15B	6	A	3.801
12B	3	D	2.808	15B	7	A	3.341
12B	4	D	3.565	15B	8	A	3.968
12B	5	D	3.988	15B	9	A	4.216
12B	6	D	3.293	15B	1	B	3.873
12B	7	D	3.518	15B	2	B	4.736
12C	1	A	4.236	15B	3	B	3.813
12C	2	A	4.233	15B	4	B	3.699
12C	3	A	4.209	15B	5	B	3.995
12C	4	A	3.249	15B	6	B	4.401
12C	5	A	4.518	15B	7	B	4.533
12C	6	A	4.155	15B	8	B	4.073
12C	7	A	3.051	15B	1	C	3.364
12C	8	A	3.738	15B	2	C	3.811
12C	9	A	4.224	15B	3	C	3.485
12C	1	B	4.026	15B	4	C	4.006
12C	2	B	3.477	15B	5	C	4.003
12C	3	B	3.813	15B	6	C	2.624
12C	4	B	4.353	15B	7	C	3.926
12C	5	B	3.669	15B	8	C	4.036
12C	6	B	4.242	15B	9	C	3.953
12C	7	B	3.711	15B	1	D	3.281
12C	8	B	4.506	15B	2	D	3.355
12C	9	B	3.033	15B	3	D	3.444
12C	10	B	3.594	15B	4	D	3.447
12C	1	C	3.960	15B	5	D	3.267
12C	2	C	3.615	15B	6	D	3.314
12C	3	C	4.062	15B	7	D	3.550
12C	4	C	3.798	15B	8	D	3.494
12C	5	C	3.972	15C	1	A	3.169
12C	6	C	4.110	15C	2	A	2.908
12C	7	C	2.748	15C	3	A	3.033
12C	8	C	4.062	15C	4	A	4.287
12C	1	D	3.639	15C	1	B	4.240
12C	2	D	3.705	15C	2	B	3.278
12C	3	D	3.408	15C	3	B	3.497
12C	4	D	3.327	15C	4	B	3.391
12C	5	D	3.249	15C	5	B	3.796
12C	6	D	3.831	15C	6	B	4.219
12C	7	D	3.240	15C	7	B	4.459
12C	8	D	4.083	15C	1	C	4.932
12C	9	D	3.444	15C	2	C	4.764
12C	10	D	3.681	15C	3	C	3.698
15B	1	A	3.801	15C	4	C	3.352
15B	2	A	4.273	15C	5	C	3.538
15B	3	A	3.870	15C	6	C	3.355
15B	4	A	3.054	15C	7	C	3.113

Appendix 1.4. Amphipod length data for the 1st set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
15C	8	C	4.077	16C	7	B	3.612
15C	1	D	3.234	16C	8	B	3.755
15C	2	D	3.589	16C	9	B	3.590
15C	3	D	3.059	16C	1	C	3.497
15C	4	D	3.391	16C	2	C	3.503
15C	5	D	3.793	16C	3	C	3.308
15C	6	D	3.166	16C	4	C	4.084
15C	7	D	3.722	16C	5	C	3.330
15C	8	D	3.056	16C	6	C	3.637
15C	9	D	3.204	16C	7	C	3.991
16B	1	A	3.196	16C	8	C	3.376
16B	2	A	3.507	16C	9	C	3.951
16B	3	A	3.547	16C	1	C	3.376
16B	4	A	4.435	16C	1	D	3.851
16B	5	A	3.302	16C	2	D	3.680
16B	6	A	3.485	16C	3	D	3.730
16B	7	A	3.581	16C	4	D	3.541
16B	8	A	3.245	16C	5	D	4.438
16B	9	A	2.920	16C	6	D	4.364
16B	1	B	3.097	16C	7	D	4.659
16B	2	B	3.838	16C	8	D	4.165
16B	3	B	3.941	16C	9	D	4.308
16B	4	B	4.510	16C	10	D	4.395
16B	5	B	3.072	16C	11	D	3.826
16B	1	C	4.078	21B	1	A	3.280
16B	2	C	3.889	21B	2	A	3.224
16B	3	C	3.805	21B	3	A	3.301
16B	4	C	3.625	21B	4	A	3.916
16B	5	C	4.134	21B	5	A	3.200
16B	6	C	3.917	21B	6	A	3.999
16B	1	D	4.280	21B	7	A	3.107
16B	2	D	4.230	21B	8	A	3.483
16B	3	D	3.699	21B	9	A	2.872
16B	4	D	3.733	21B	10	A	3.265
16B	5	D	3.764	21B	1	B	3.283
16B	6	D	3.864	21B	2	B	3.319
16C	1	A	4.261	21B	3	B	3.030
16C	2	A	3.494	21B	4	B	3.781
16C	3	A	4.469	21B	5	B	4.160
16C	4	A	3.901	21B	6	B	4.178
16C	5	A	3.929	21B	7	B	2.809
16C	6	A	2.770	21B	8	B	3.775
16C	1	B	4.581	21B	9	B	3.856
16C	2	B	3.327	21B	1	C	3.480
16C	3	B	3.777	21B	2	C	3.161
16C	4	B	3.907	21B	3	C	3.808
16C	5	B	3.578	21B	4	C	3.579
16C	6	B	4.009	21B	5	C	4.056

Appendix 1.4. Amphipod length data for the 1st set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
21B	6	C	3.501	25B	1	A	2.579
21B	7	C	3.960	25B	2	A	4.374
21B	8	C	3.579	25B	3	A	3.433
21B	1	D	3.611	25B	4	A	3.347
21B	2	D	2.887	25B	5	A	3.735
21B	3	D	3.140	25B	6	A	2.955
21B	4	D	3.632	25B	1	B	3.777
21B	5	D	2.565	25B	2	B	3.726
21B	6	D	3.543	25B	3	B	3.840
21B	7	D	3.811	25B	4	B	3.735
21B	8	D	3.012	25B	5	B	4.225
21B	9	D	3.537	25B	6	B	4.237
21B	10	D	3.254	25B	7	B	4.129
21C	1	A	4.595	25B	1	C	3.816
21C	2	A	3.895	25B	2	C	2.854
21C	3	A	4.476	25B	3	C	3.675
21C	4	A	3.397	25B	4	C	4.094
21C	5	A	4.512	25B	5	C	3.923
21C	6	A	3.343	25B	1	D	3.837
21C	7	A	4.509	25B	2	D	2.561
21C	8	A	3.850	25B	3	D	3.565
21C	9	A	3.808	25B	4	D	3.430
21C	1	B	3.069	25B	5	D	2.976
21C	2	B	3.358	25C	1	A	2.952
21C	3	B	2.863	25C	2	A	3.120
21C	4	B	3.295	25C	3	A	3.953
21C	5	B	3.069	25C	4	A	3.831
21C	6	B	3.671	25C	5	A	4.052
21C	7	B	3.110	25C	6	A	3.834
21C	8	B	3.376	25C	7	A	3.729
21C	1	C	3.865	25C	8	A	3.093
21C	2	C	4.539	25C	1	B	5.053
21C	3	C	4.109	25C	2	B	3.920
21C	4	C	4.366	25C	1	C	3.547
21C	5	C	4.921	25C	2	C	3.487
21C	6	C	2.920	25C	3	C	3.469
21C	7	C	3.901	25C	4	C	3.299
21C	8	C	3.620	25C	5	C	3.215
21C	1	D	4.047	25C	6	C	3.681
21C	2	D	4.273	25C	7	C	3.356
21C	3	D	3.373	25C	8	C	3.108
21C	4	D	4.545	25C	9	C	3.571
21C	5	D	3.987	25C	10	C	3.448
21C	6	D	3.957	25C	1	D	3.645
21C	7	D	4.643	25C	2	D	4.494
21C	8	D	4.094	25C	3	D	3.344
21C	9	D	4.050	25C	4	D	3.538
21C	10	D	4.088	25C	5	D	3.729

Appendix 1.4. Amphipod length data for the 1st set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
25C	6	D	3.266	26C	10	A	2.829
25C	7	D	3.648	26C	1	B	2.764
25C	8	D	3.887	26C	2	B	3.267
25C	9	D	3.953	26C	3	B	3.084
26B	1	A	3.090	26C	4	B	2.713
26B	2	A	3.837	26C	5	B	2.875
26B	3	A	3.750	26C	6	B	3.171
26B	4	A	3.642	26C	7	B	3.006
26B	5	A	4.744	26C	1	C	2.761
26B	6	A	4.735	26C	2	C	2.686
26B	7	A	4.163	26C	3	C	2.731
26B	8	A	3.319	26C	4	C	2.883
26B	9	A	4.072	26C	5	C	2.632
26B	10	A	4.603	26C	6	C	2.680
26B	1	B	3.883	26C	1	D	3.204
26B	2	B	4.253	26C	2	D	3.012
26B	3	B	2.313	26C	3	D	2.665
26B	4	B	3.762	26C	4	D	2.620
26B	5	B	3.139	26C	5	D	2.593
26B	6	B	3.536	26C	6	D	2.764
26B	7	B	3.825	26C	7	D	2.964
26B	8	B	4.184	26C	8	D	3.054
26B	1	C	3.072	26C	9	D	2.958
26B	2	C	2.958	26C	10	D	2.255
26B	3	C	3.293	FLOR	1	A	2.734
26B	4	C	2.922	FLOR	2	A	4.479
26B	5	C	3.338	FLOR	3	A	3.796
26B	6	C	3.039	FLOR	4	A	3.362
26B	7	C	3.533	FLOR	5	A	4.323
26B	8	C	2.961	FLOR	6	A	3.401
26B	9	C	3.003	FLOR	7	A	3.826
26B	1	D	2.946	FLOR	1	B	4.156
26B	2	D	3.740	FLOR	2	B	3.087
26B	3	D	3.434	FLOR	3	B	3.237
26B	4	D	3.219	FLOR	4	B	3.278
26B	5	D	3.084	FLOR	5	B	4.587
26B	6	D	2.931	FLOR	6	B	2.455
26B	7	D	2.925	FLOR	7	B	3.332
26B	8	D	3.344	FLOR	8	B	3.434
26C	1	A	2.701	FLOR	1	C	2.731
26C	2	A	3.018	FLOR	2	C	1.338
26C	3	A	3.039	FLOR	3	C	1.976
26C	4	A	2.832	FLOR	4	C	3.683
26C	5	A	2.659	FLOR	5	C	3.668
26C	6	A	3.341	FLOR	6	C	2.973
26C	7	A	3.063	FLOR	7	C	1.868
26C	8	A	3.012	FLOR	1	D	4.341
26C	9	A	3.126	FLOR	2	D	3.790

Appendix 1.4. Amphipod length data for the 1st set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
FLOR	3	D	3.644	FLOR	7	D	3.976
FLOR	4	D	4.072	FLOR	8	D	4.231
FLOR	5	D	2.578	FLOR	9	D	1.967
FLOR	6	D	4.760				

Appendix 1.5. Amphipod length data for the 2nd set of samples. Replication (Rep), Animal (individual animal number), and length (mean length for individual animal).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
ARCH	1	NA	1.339	2C	6	B	3.066
ARCH	2	NA	1.369	2C	7	B	3.475
ARCH	3	NA	1.193	2C	8	B	3.200
ARCH	4	NA	1.178	2C	9	B	2.755
ARCH	5	NA	1.101	2C	10	B	3.956
ARCH	6	NA	1.107	2C	1	C	2.848
ARCH	7	NA	1.021	2C	2	C	3.938
ARCH	8	NA	1.056	2C	3	C	4.109
ARCH	9	NA	1.134	2C	4	C	3.884
ARCH	10	NA	1.196	2C	5	C	3.514
2B	1	A	4.177	2C	6	C	3.804
2B	2	A	3.556	2C	7	C	4.046
2B	3	A	3.890	2C	1	D	4.593
2B	4	A	4.572	2C	2	D	3.093
2B	5	A	4.001	2C	3	D	2.352
2B	6	A	3.920	2C	4	D	4.001
2B	7	A	4.467	2C	5	D	3.968
2B	8	A	4.195	2C	6	D	4.443
2B	1	B	4.461	2C	7	D	3.117
2B	2	B	4.491	2C	8	D	4.234
2B	3	B	3.729	2C	9	D	4.243
2B	4	B	4.246	4B	1	A	3.616
2B	5	B	4.622	4B	2	A	4.485
2B	6	B	3.502	4B	3	A	4.467
2B	7	B	4.718	4B	4	A	3.174
2B	8	B	3.421	4B	1	B	3.337
2B	1	C	5.184	4B	2	B	3.243
2B	2	C	3.980	4B	3	B	3.295
2B	3	C	5.558	4B	4	B	2.579
2B	4	C	4.213	4B	5	B	3.391
2B	1	D	2.874	4B	6	B	3.343
2B	2	D	4.548	4B	7	B	4.148
2B	3	D	3.027	4B	8	B	2.841
2B	4	D	3.711	4B	1	C	3.415
2B	5	D	3.568	4B	2	C	3.457
2B	6	D	4.040	4B	3	C	3.270
2B	7	D	3.777	4B	4	C	3.682
2B	8	D	3.436	4B	5	C	3.944
2C	1	A	3.108	4B	6	C	2.291
2C	2	A	2.898	4B	7	C	3.433
2C	3	A	2.737	4B	8	C	3.388
2C	4	A	2.880	4B	9	C	4.413
2C	5	A	3.102	4B	1	D	2.600
2C	1	B	3.096	4B	2	D	2.444
2C	2	B	3.442	4B	3	D	3.860
2C	3	B	3.359	4B	4	D	4.205
2C	4	B	3.403	4B	5	D	3.240
2C	5	B	2.764	4B	6	D	2.850

Appendix 1.5. Amphipod length data for the 2nd set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
4B	7	D	3.502	6B	6	C	4.360
4B	8	D	3.213	6B	7	C	3.045
4B	9	D	2.714	6B	8	C	3.200
4B	10	D	3.821	6B	9	C	3.242
4B	11	D	3.478	6B	10	C	3.831
4B	12	D	2.868	6B	1	D	2.943
4B	13	D	2.922	6B	2	D	3.311
4B	14	D	3.785	6B	3	D	3.935
4C	1	B	3.024	6B	4	D	4.634
4C	2	B	2.744	6B	5	D	3.553
4C	3	B	2.594	6B	6	D	2.994
4C	4	B	3.661	6B	7	D	3.335
4C	5	B	2.802	6B	8	D	3.777
4C	6	B	3.195	6C	1	A	3.568
4C	7	B	2.934	6C	2	A	3.819
4C	8	B	3.556	6C	3	A	2.943
4C	9	B	3.562	6C	4	A	4.204
4C	10	B	3.234	6C	5	A	4.336
4C	11	B	3.439	6C	6	A	5.462
4C	1	C	3.093	6C	7	A	3.027
4C	2	C	3.030	6C	8	A	4.608
4C	3	C	4.269	6C	9	A	4.019
4C	4	C	3.036	6C	1	B	4.790
4C	5	C	3.039	6C	2	B	4.775
4C	6	C	3.986	6C	3	B	4.183
4C	7	C	3.830	6C	4	B	3.870
4C	8	C	3.144	6C	5	B	4.688
4C	1	D	3.493	6C	6	B	4.760
4C	2	D	3.758	6C	7	B	4.617
4C	3	D	4.323	6C	8	B	3.855
4C	4	D	3.721	6C	1	C	3.583
4C	5	D	3.427	6C	2	C	4.682
4C	6	D	3.195	6C	3	C	3.439
4C	7	D	3.090	6C	4	C	3.317
6B	1	A	3.317	6C	5	C	4.969
6B	2	A	3.508	6C	6	C	3.601
6B	3	A	3.499	6C	7	C	3.589
6B	4	A	3.326	6C	8	C	4.115
6B	5	A	3.132	6C	1	D	3.765
6B	6	A	3.493	6C	2	D	3.911
6B	7	A	2.949	6C	3	D	3.622
6B	1	B	3.024	6C	4	D	3.941
6B	2	B	3.908	6C	5	D	4.318
6B	1	C	3.132	6C	6	D	4.464
6B	2	C	3.636	6C	7	D	4.085
6B	3	C	4.013	6C	8	D	3.657
6B	4	C	4.443	7B	1	A	3.886
6B	5	C	3.741	7B	2	A	3.844

Appendix 1.5. Amphipod length data for the 2nd set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
7B	3	A	3.605	7C	2	B	3.593
7B	4	A	3.725	7C	3	B	3.889
7B	5	A	3.635	7C	4	B	3.859
7B	6	A	3.695	7C	5	B	3.546
7B	7	A	3.361	7C	6	B	4.172
7B	8	A	4.294	7C	7	B	4.232
7B	9	A	3.537	7C	8	B	3.304
7B	10	A	4.127	7C	9	B	3.895
7B	1	B	3.263	7C	10	B	4.348
7B	2	B	3.185	7C	1	C	3.507
7B	3	B	3.835	7C	2	C	3.450
7B	4	B	3.447	7C	3	C	4.578
7B	5	B	3.263	7C	4	C	3.725
7B	6	B	3.587	7C	5	C	3.701
7B	7	B	4.262	7C	6	C	3.435
7B	8	B	3.832	7C	7	C	3.656
7B	9	B	3.158	7C	8	C	3.534
7B	10	B	3.641	7C	9	C	3.898
7B	1	C	3.549	7C	1	D	3.602
7B	2	C	3.531	7C	2	D	3.743
7B	3	C	3.531	7C	3	D	4.005
7B	4	C	3.140	7C	4	D	3.925
7B	5	C	3.087	7C	5	D	3.811
7B	6	C	3.084	7C	6	D	3.671
7B	7	C	3.486	7C	7	D	3.352
7B	8	C	3.110	7C	8	D	3.307
7B	9	C	4.411	7C	9	D	3.477
7B	10	C	3.570	7C	10	D	3.632
7B	1	D	4.470	7C	11	D	1.813
7B	2	D	4.136	9B	1	A	4.027
7B	3	D	4.238	9B	2	A	4.675
7B	4	D	4.050	9B	3	A	4.033
7B	5	D	3.543	9B	4	A	3.801
7B	6	D	3.707	9B	5	A	3.178
7B	7	D	3.638	9B	6	A	3.587
7B	8	D	3.948	9B	7	A	3.443
7B	9	D	3.543	9B	1	B	3.301
7B	10	D	3.590	9B	2	B	3.398
7C	1	A	4.405	9B	3	B	5.358
7C	2	A	4.160	9B	4	B	4.527
7C	3	A	3.283	9B	5	B	3.346
7C	4	A	3.659	9B	1	C	3.214
7C	5	A	3.468	9B	2	C	3.455
7C	6	A	3.090	9B	3	C	3.705
7C	7	A	4.005	9B	4	C	3.844
7C	8	A	3.361	9B	5	C	3.870
7C	9	A	3.987	9B	6	C	3.654
7C	1	B	4.178	9B	7	C	3.843

Appendix 1.5. Amphipod length data for the 2nd set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
9B	8	C	3.772	13B	1	C	4.581
9B	9	C	3.352	13B	2	C	3.388
9B	10	C	3.566	13B	3	C	4.079
9B	11	C	3.461	13B	4	C	3.308
9B	1	D	3.662	13B	5	D	3.006
9B	2	D	3.759	13B	1	D	2.766
9B	3	D	2.822	13B	2	D	3.970
9B	4	D	3.304	13B	3	D	3.635
9B	5	D	4.416	13B	4	D	3.701
9B	6	D	3.753	13C	1	A	3.968
9B	7	D	3.699	13C	2	A	3.553
9B	8	D	3.361	13C	3	A	3.460
9C	1	A	2.913	13C	1	B	3.364
9C	2	A	2.976	13C	2	B	3.290
9C	3	A	3.666	13C	3	B	3.905
9C	4	A	4.283	13C	4	B	4.138
9C	1	B	4.093	13C	5	B	3.759
9C	2	B	3.843	13C	6	B	3.977
9C	3	B	3.374	13C	1	C	2.940
9C	4	B	4.202	13C	2	C	3.448
9C	5	B	4.081	13C	3	C	2.851
9C	6	B	3.708	13C	4	C	4.643
9C	7	B	4.280	13C	5	C	3.565
9C	8	B	4.018	13C	6	C	2.737
9C	1	C	3.072	13C	1	D	3.478
9C	2	C	3.229	13C	2	D	3.565
9C	3	C	3.334	13C	3	D	3.451
9C	4	C	3.982	14B	1	A	3.092
9C	5	C	3.681	14B	2	A	3.693
9C	6	C	3.509	14B	3	A	3.503
9C	7	C	3.178	14B	4	A	3.235
9C	8	C	3.340	14B	5	A	4.714
9C	9	C	3.566	14B	6	A	3.271
9C	10	C	3.692	14B	7	A	3.024
9C	11	C	3.162	14B	1	B	4.211
9C	12	C	2.825	14B	2	B	3.429
9C	13	C	3.289	14B	3	B	4.274
9C	14	C	3.656	14B	4	B	4.482
9C	15	C	4.075	14B	5	B	3.786
9C	16	C	4.256	14B	6	B	4.092
9C	1	D	4.100	14B	7	B	5.723
9C	2	D	3.991	14B	1	C	3.542
9C	3	D	3.256	14B	2	C	3.408
9C	4	D	4.142	14B	3	C	3.780
13B	1	A	5.304	14B	4	C	2.872
13B	2	A	4.062	14B	5	C	4.452
13B	1	B	4.652	14B	6	C	4.036
13B	2	B	3.870	14B	1	D	3.934

Appendix 1.5. Amphipod length data for the 2nd set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
14B	2	D	3.173	18B	3	D	3.534
14B	3	D	3.756	18B	4	D	3.618
14B	4	D	3.863	18B	5	D	3.693
14B	5	D	4.821	18C	1	A	3.608
14B	6	D	4.036	18C	2	A	3.310
14B	7	D	3.863	18C	3	A	3.841
14C	1	A	3.434	18C	4	A	2.696
14C	2	A	3.066	18C	5	A	3.412
14C	3	A	4.009	18C	6	A	4.169
14C	4	A	3.268	18C	7	A	3.793
14C	5	A	4.053	18C	8	A	3.552
14C	6	A	3.426	18C	9	A	2.767
14C	1	B	2.568	18C	1	B	2.857
14C	2	B	3.009	18C	2	B	3.823
14C	3	B	2.640	18C	3	B	4.601
14C	4	B	2.631	18C	4	B	3.626
14C	5	B	3.765	18C	5	B	3.304
14C	6	B	4.054	18C	1	C	3.444
14C	1	C	3.527	18C	2	C	3.578
14C	2	C	4.161	18C	3	C	4.178
14C	3	C	3.134	18C	4	C	2.994
14C	4	C	3.946	18C	5	C	3.414
14C	1	D	3.934	18C	6	C	3.868
14C	2	D	3.845	18C	7	C	2.896
14C	3	D	4.077	18C	8	C	3.728
18B	1	A	3.816	18C	9	C	3.155
18B	2	A	3.375	18C	10	C	3.364
18B	3	A	2.355	18C	1	D	3.904
18B	4	A	4.167	19B	1	A	4.054
18B	5	A	3.978	19B	2	A	3.348
18B	6	A	3.000	19B	3	A	3.872
18B	7	A	2.559	19B	4	A	3.339
18B	1	B	4.803	19B	5	A	3.304
18B	2	B	3.636	19B	6	A	3.167
18B	3	B	2.898	19B	7	A	3.146
18B	4	B	3.681	19B	1	B	3.348
18B	5	B	3.738	19B	2	B	4.214
18B	6	B	4.503	19B	3	B	2.450
18B	7	B	4.020	19B	4	B	3.455
18B	1	C	3.618	19B	5	B	2.938
18B	2	C	2.664	19B	6	B	3.923
18B	3	C	3.711	19B	7	B	3.506
18B	4	C	3.579	19B	8	B	3.726
18B	5	C	3.570	19B	9	B	3.173
18B	6	C	2.946	19B	1	C	3.774
18B	7	C	2.928	19B	2	C	3.851
18B	1	D	4.065	19B	3	C	3.378
18B	2	D	4.434	19B	4	C	3.295

Appendix 1.5. Amphipod length data for the 2nd set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
19B	5	C	2.818	20B	7	A	3.454
19B	6	C	3.711	20B	8	A	2.940
19B	7	C	2.970	20B	1	B	3.433
19B	8	C	3.205	20B	2	B	3.663
19B	1	D	3.009	20B	3	B	3.577
19B	2	D	3.304	20B	4	B	3.837
19B	3	D	3.319	20B	5	B	3.269
19B	4	D	3.015	20B	6	B	3.317
19B	5	D	2.774	20B	7	B	3.768
19B	6	D	3.089	20B	8	B	3.281
19B	7	D	3.545	20B	9	B	4.132
19B	8	D	2.616	20B	10	B	3.995
19B	9	D	2.836	20B	1	C	3.122
19B	10	D	2.917	20B	2	C	2.510
19C	1	A	3.893	20B	3	C	2.854
19C	2	A	3.792	20B	4	C	4.183
19C	3	A	3.238	20B	5	C	3.155
19C	4	A	2.929	20B	6	C	3.442
19C	5	A	3.581	20B	7	C	3.380
19C	6	A	2.515	20B	8	C	3.353
19C	7	A	3.512	20B	1	D	2.582
19C	8	A	4.137	20B	2	D	2.872
19C	1	B	3.631	20B	3	D	2.883
19C	2	B	3.777	20B	4	D	3.433
19C	3	B	2.997	20B	5	D	3.738
19C	4	B	3.176	20B	6	D	3.030
19C	5	B	3.875	20C	1	A	3.669
19C	6	B	2.979	20C	2	A	3.807
19C	1	C	3.646	20C	3	A	3.454
19C	2	C	3.256	20C	4	A	4.138
19C	3	C	3.435	20C	5	A	3.403
19C	4	C	3.955	20C	6	A	3.750
19C	5	C	3.438	20C	7	A	3.634
19C	6	C	3.307	20C	8	A	3.030
19C	7	C	3.149	20C	9	A	3.466
19C	8	C	3.720	20C	1	B	3.370
19C	1	D	2.920	20C	2	B	3.379
19C	2	D	3.845	20C	3	B	2.794
19C	3	D	3.366	20C	4	B	3.466
19C	4	D	3.494	20C	5	B	3.364
19C	5	D	3.173	20C	6	B	3.547
19C	6	D	3.485	20C	7	B	2.749
20B	1	A	3.630	20C	8	B	3.484
20B	2	A	3.580	20C	9	B	3.457
20B	3	A	4.004	20C	1	C	3.472
20B	4	A	3.711	20C	2	C	2.994
20B	5	A	3.834	20C	3	C	3.374
20B	6	A	3.884	20C	4	C	3.320

Appendix 1.5. Amphipod length data for the 2nd set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
20C	5	C	3.350	22B	6	D	3.873
20C	6	C	3.418	22C	1	A	3.914
20C	7	C	3.143	22C	2	A	4.025
20C	8	C	3.021	22C	3	A	4.679
20C	9	C	3.780	22C	1	B	3.335
20C	1	D	2.979	22C	2	B	3.813
20C	2	D	2.964	22C	3	B	3.126
20C	3	D	3.230	22C	4	B	3.063
20C	4	D	3.185	22C	5	B	3.598
20C	5	D	3.388	22C	6	B	3.601
20C	6	D	2.522	22C	7	B	2.674
20C	7	D	3.182	22C	8	B	3.935
20C	8	D	3.015	22C	1	C	3.457
20C	9	D	3.221	22C	2	C	3.998
20C	10	D	2.564	22C	3	C	3.427
22B	1	A	3.586	22C	4	C	2.755
22B	2	A	3.708	22C	5	C	3.391
22B	3	A	3.786	22C	1	D	3.610
22B	4	A	4.004	22C	2	D	3.227
22B	5	A	3.063	22C	3	D	4.144
22B	6	A	3.550	22C	4	D	3.436
22B	7	A	3.350	22C	5	D	4.856
22B	8	A	4.121	22C	6	D	3.311
22B	9	A	3.415	22C	7	D	4.252
22B	10	A	4.778	24B	1	A	3.925
22B	1	B	5.253	24B	2	A	2.958
22B	2	B	4.351	24B	3	A	3.886
22B	3	B	3.666	24B	4	A	3.617
22B	4	B	3.732	24B	5	A	3.533
22B	5	B	3.281	24B	6	A	3.781
22B	6	B	3.173	24B	7	A	3.434
22B	7	B	3.885	24B	8	A	4.302
22B	8	B	3.290	24B	1	B	2.263
22B	9	B	2.848	24B	2	B	3.165
22B	1	C	3.188	24B	3	B	3.820
22B	2	C	3.639	24B	4	B	4.159
22B	3	C	4.760	24B	5	B	4.108
22B	4	C	3.726	24B	6	B	3.536
22B	5	C	4.527	24B	7	B	4.054
22B	6	C	4.258	24B	8	B	3.599
22B	7	C	4.978	24B	9	B	3.329
22B	8	C	2.934	24B	1	C	3.293
22B	9	C	3.729	24B	2	C	4.045
22B	1	D	3.595	24B	3	C	4.362
22B	2	D	3.753	24B	4	C	2.931
22B	3	D	3.556	24B	5	C	3.012
22B	4	D	3.917	24B	6	C	3.707
22B	5	D	3.636	24B	7	C	3.922

Appendix 1.5. Amphipod length data for the 2nd set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
24B	8	C	3.189	SCB	5	C	2.784
24B	9	C	3.257	SCB	6	C	2.651
24B	1	D	3.862	SCB	7	C	3.661
24B	2	D	3.877	SCB	1	D	3.605
24B	3	D	3.563	SCB	2	D	3.708
24B	4	D	3.931	SCB	3	D	4.795
24B	5	D	3.054	SCB	4	D	3.343
24B	6	D	3.060	SCB	5	D	3.352
24B	7	D	3.653	SCB	6	D	2.955
24B	8	D	3.722	SCC	1	A	3.373
24B	9	D	4.311	SCC	2	A	2.286
24C	1	A	3.877	SCC	3	A	2.506
24C	2	A	3.750	SCC	4	A	2.973
24C	3	A	4.007	SCC	5	A	2.678
24C	4	A	3.490	SCC	6	A	3.051
24C	5	A	2.985	SCC	7	A	3.331
24C	6	A	3.414	SCC	8	A	3.238
24C	7	A	3.481	SCC	1	B	3.259
24C	8	A	3.980	SCC	2	B	2.955
24C	1	B	2.822	SCC	3	B	3.346
24C	2	B	3.820	SCC	4	B	3.563
24C	3	B	3.841	SCC	5	B	3.518
24C	4	B	4.542	SCC	6	B	2.952
24C	5	B	3.611	SCC	7	B	3.111
24C	6	B	4.836	SCC	8	B	2.961
24C	1	C	3.157	SCC	1	C	2.789
24C	2	C	3.859	SCC	2	C	2.584
24C	3	C	3.505	SCC	3	C	2.732
24C	4	C	3.832	SCC	4	C	2.753
24C	5	C	3.756	SCC	5	C	2.937
24C	6	C	3.139	SCC	6	C	2.861
24C	1	D	4.001	SCC	7	C	2.855
24C	2	D	5.571	SCC	8	C	2.922
24C	3	D	3.387	SCC	9	C	3.352
24C	4	D	4.083	SCC	10	C	2.404
SCB	1	A	3.794	SCC	1	D	2.725
SCB	2	A	3.490	SCC	2	D	3.275
SCB	3	A	3.758	SCC	3	D	3.359
SCB	4	A	3.171	SCC	4	D	3.741
SCB	1	B	3.748	SCC	5	D	3.648
SCB	2	B	3.736	SCC	6	D	2.537
SCB	3	B	2.994	SCC	7	D	2.523
SCB	4	B	3.239	SCC	8	D	3.143
SCB	5	B	3.393	SCC	9	D	3.648
SCB	1	C	3.325	FLOR	1	A	2.334
SCB	2	C	2.933	FLOR	2	A	2.522
SCB	3	C	3.319	FLOR	3	A	2.256
SCB	4	C	3.583	FLOR	4	A	2.561

Appendix 1.5. Amphipod length data for the 2nd set of sediment samples (continued).

Sample	Animal	Rep	Length	Sample	Animal	Rep	Length
FLOR	5	A	2.624	FLOR	4	C	3.606
FLOR	6	A	2.546	FLOR	5	C	2.444
FLOR	7	A	1.743	FLOR	6	C	2.916
FLOR	8	A	1.960	FLOR	7	C	2.866
FLOR	9	A	4.070	FLOR	8	C	2.761
FLOR	1	B	2.394	FLOR	1	D	2.113
FLOR	2	B	1.978	FLOR	2	D	1.909
FLOR	3	B	2.913	FLOR	3	D	2.576
FLOR	4	B	2.949	FLOR	4	D	1.790
FLOR	5	B	2.650	FLOR	5	D	2.283
FLOR	6	B	2.850	FLOR	6	D	2.360
FLOR	7	B	2.901	FLOR	7	D	2.522
FLOR	8	B	3.436	FLOR	8	D	3.347
FLOR	9	B	2.268	FLOR	9	D	2.668
FLOR	10	B	2.531	FLOR	10	D	2.059
FLOR	1	C	3.320	FLOR	11	D	1.981
FLOR	2	C	2.328				
FLOR	3	C	3.230				

Appendix 1.6. Amphipod maturation and survival data for the 1st set of samples. Replication (Rep), number of amphipods recovered (Recov), and number of males recovered (Males).

Sample	Rep	Recov	Males	Sample	Rep	Recov	Males
01B	1	8	3	12C	1	9	4
01B	2	10	3	12C	2	10	2
01B	3	9	5	12C	3	8	2
01B	4	9	3	12C	4	10	4
01C	1	5	1	15B	1	9	3
01C	2	6	0	15B	2	8	6
01C	3	7	1	15B	3	9	6
01C	4	6	2	15B	4	10	3
03B	1	4	1	15C	1	7	1
03B	2	8	3	15C	2	7	2
03B	3	11	6	15C	3	8	3
03B	4	8	5	15C	4	9	5
05B	1	9	2	16B	1	9	2
05B	2	9	6	16B	2	5	2
05B	3	7	4	16B	3	6	3
05B	4	6	2	16B	4	6	3
05C	1	7	1	16C	1	6	1
05C	2	6	1	16C	2	9	1
05C	3	9	3	16C	3	9	5
05C	4	9	2	16C	4	11	4
08B	1	10	5	21B	1	10	5
08B	2	10	2	21B	2	9	4
08B	3	8	3	21B	3	9	4
08B	4	11	6	21B	4	10	7
08C	1	9	3	21C	1	9	5
08C	2	6	3	21C	2	8	3
08C	3	8	1	21C	3	8	5
08C	4	9	3	21C	4	10	5
10B	1	10	5	25B	1	6	1
10B	2	9	2	25B	2	7	2
10B	3	7	6	25B	3	5	0
10B	4	10	0	25B	4	4	2
10C	1	6	3	25C	1	8	2
10C	2	6	1	25C	2	2	1
10C	3	9	3	25C	3	10	1
10C	4	8	3	25C	4	9	3
11B	1	7	2	26B	1	10	5
11B	2	9	3	26B	2	8	2
11B	3	9	3	26B	3	9	5
11B	4	9	7	26B	4	8	3
11C	1	7	1	26C	1	10	7
11C	2	5	4	26C	2	7	2
11C	3	6	1	26C	3	6	4
11C	4	5	1	26C	4	10	3
12B	1	9	3	FLB	1	6	2
12B	2	9	3	FLB	2	8	5
12B	3	7	2	FLB	3	7	2
12B	4	7	3	FLB	4	9	2

Appendix 1.7. Amphipod maturation and survival data for the 2nd set of samples. Replication (Rep), number of amphipods recovered (Recov), and number of males recovered (Males).

Sample	Rep	Recov	Males	Sample	Rep	Recov	Males
02B	1	8	4	14B	1	7	1
02B	2	8	2	14B	2	7	2
02B	3	4	1	14B	3	6	3
02B	4	8	2	14B	4	6	2
02C	1	5	2	14C	1	6	1
02C	2	10	4	14C	2	6	1
02C	3	7	2	14C	3	4	3
02C	4	9	6	14C	4	3	2
04B	1	4	3	18B	1	7	2
04B	2	8	2	18B	2	7	4
04B	3	9	1	18B	3	7	1
04B	4	14	5	18B	4	5	5
04C	1	0	0	18C	1	9	3
04C	2	11	1	18C	2	5	1
04C	3	8	2	18C	3	10	3
04C	4	7	1	18C	4	1	0
06B	1	7	1	19B	1	7	3
06B	2	2	1	19B	2	9	7
06B	3	10	1	19B	3	8	0
06B	4	6	2	19B	4	10	4
06C	1	9	5	19C	1	8	3
06C	2	8	4	19C	2	6	3
06C	3	8	4	19C	3	8	2
06C	4	8	5	19C	4	6	1
07B	1	10	2	20B	1	8	2
07B	2	10	7	20B	2	10	1
07B	3	10	4	20B	3	8	1
07B	4	10	4	20B	4	6	0
07C	1	9	3	20C	1	9	4
07C	2	10	3	20C	2	9	0
07C	3	9	3	20C	3	9	4
07C	4	11	5	20C	4	10	2
09B	1	7	4	22B	1	10	2
09B	2	5	2	22B	2	9	2
09B	3	11	3	22B	3	9	2
09B	4	8	4	22B	4	6	2
09C	1	4	2	22C	1	3	2
09C	2	8	3	22C	2	8	4
09C	3	16	7	22C	3	5	0
09C	4	4	0	22C	4	7	3
13B	1	2	0	24B	1	8	3
13B	2	2	1	24B	2	9	2
13B	3	5	0	24B	3	9	4
13B	4	4	1	24B	4	9	3
13C	1	3	2	24C	1	8	3
13C	2	6	2	24C	2	5	4
13C	3	6	2	24C	3	6	6
13C	4	3	2	24C	4	4	2

Appendix 1.7. (Continued)

Sample	Rep	Recov	Males	Sample	Rep	Recov	Males
SCB	1	4	0	FLC	1	9	1
SCB	2	5	0	FLC	2	10	0
SCB	3	7	1	FLC	3	8	1
SCB	4	6	2	FLC	4	11	0
SCC	1	8	2				
SCC	2	8	3				
SCC	3	10	2				
SCC	4	9	4				

Appendix 1.8. Concentrations of acid volatile sulfide (μmoles/g) and simultaneously extracted metals (SEM in ug/g dry weight) and the sum of the molar concentration of SEMs and the sum of the molar concentration of SEM divided by the molar concentration of AVS for the Upper Mississippi River sediment samples.

Sample	AVS	Cd	Cu	Ni	Pb	Zn	∑SEM	∑SEM/AVS
1B	0.19	0.058	1.1	1.2	5.2	11	0.232	1.22
1C	0.005	2.1	0.67	0.76	4.3	7	0.170	17.00
2B	1.9	0.39	4.7	4.6	7.7	26	0.591	0.31
2C	3.1	0.91	8.4	7.6	13.0	47	1.051	0.34
3B	2.1	0.68	7.3	6.1	11.0	35	0.813	0.39
4B	9.4	1.5	13.0	9.9	26.0	73	1.629	0.17
4C	16.0	3.0	20.0	15.0	53.0	118	2.658	0.17
5B	1.9	0.21	3.4	3.0	4.8	15	0.359	0.19
5C	16.0	0.63	6.3	9.4	16.0	45	1.030	0.06
6B	0.8	0.062	0.85	1.5	2.1	7.1	0.158	0.20
6C	2.9	0.12	2.0	2.7	4.2	12	0.282	0.10
7B	1.4	0.2	2.7	4.6	4.6	15	0.374	0.27
7C	8.7	0.43	5.3	6.6	12.0	35	0.793	0.09
8B	0.6	0.14	2.4	2.6	4.1	12	0.287	0.48
8C	2.2	0.28	4.1	4.6	6.9	20	0.485	0.22
9B	3.8	0.3	4.4	4.9	8.4	24	0.563	0.15
9C	3.8	0.38	5.2	5.4	11.0	31	0.705	0.19
10B	4.8	0.17	3.7	3.2	6.0	18	0.419	0.09
10C	63.0	0.74	11.0	10.0	26.0	68	1.516	0.02
11B	1.1	0.13	2.5	3.2	5.8	16	0.368	0.33
11C	6.5	0.18	2.8	3.8	7.1	17	0.405	0.06
12B	3.8	0.43	4.8	4.8	26.0	137	2.382	0.63
12C	5.9	0.45	4.5	4.1	27.0	143	2.462	0.42
13B	3.1	0.14	2.3	3.2	6.8	22	0.461	0.15
13C	5.2	0.3	4.4	4.5	14.0	48	0.950	0.18
14B	3.5	0.14	2.8	3.3	6.1	19	0.422	0.12
14C	18.0	0.41	6.8	6.6	18.0	51	1.090	0.06
15B	3.6	0.2	4.0	3.9	9.9	31	0.653	0.18
15C	3.7	0.34	5.3	4.5	14.0	44	0.904	0.24
16B	2.0	0.14	2.5	3.1	5.8	20	0.427	0.21
16C	3.5	0.31	2.7	2.9	8.0	30	0.592	0.17

Appendix 1.8. (continued)

Sample	AVS	Cd	Cu	Ni	Pb	Zn	∑SEM	∑SEM/AVS
18B	2.8	0.17	2.6	3.1	5.6	20	0.428	0.15
18C	7.1	0.36	3.6	3.4	9.2	33	0.667	0.09
19B	4.1	0.28	4.3	5.0	8.8	27	0.611	0.15
19C	5.1	0.28	5.4	5.3	11.0	30	0.690	0.14
20B	0.1	0.028	0.49	1.4	1.7	5.6	0.126	0.79
20C	1.8	0.15	2.4	2.6	5.3	15	0.338	0.19
21B	1.0	0.095	2.1	2.2	4.2	12	0.275	0.28
21C	2.4	0.16	3.6	3.6	5.9	16	0.393	0.16
22B	1.8	0.12	2.8	2.8	6.3	16	0.368	0.20
22C	10.0	0.4	4.9	5.1	13.0	38	0.812	0.08
24B	0.81	0.091	2.5	2.8	4.3	11	0.277	0.34
24C	3.2	0.28	4.3	4.4	8.9	26	0.586	0.18
25B	1.2	0.19	4.6	4.0	7.7	18	0.455	0.38
25C	2.9	0.19	3.8	3.8	8.3	17	0.426	0.15
26B	2.4	0.27	7.1	6.0	15.0	25	0.671	0.28
26C	1.7	0.15	3.4	5.0	11.0	19	0.484	0.28
SCB	1.7	0.31	9.0	3.4	17.0	35	0.820	0.48
SCC	5.5	0.54	14.0	4.9	30.0	53	1.264	0.23

Appendix 1.9. Concentrations (µg/g dry weight) of organochlorine pesticides (OCs) in Upper Mississippi River sediments.

POOL	Chlordane	Dieldrin	DDE	DDD	DDT
1B	0.001	ND[1]	0.0004	0.0005	ND
1C	ND	ND	ND	ND	ND
2B	0.001	0.0003	ND	0.0016	0.0002
2C	ND	ND	0.0520	0.0790	ND
3B	ND	0.0003	0.0011	0.0038	0.0002
4B	0.002	0.0005	0.0010	0.0019	ND
4C	ND	ND	ND	ND	ND
5B	ND	ND	0.0001	0.0001	ND
5C	ND	ND	ND	ND	ND
6B	ND	ND	0.0001	0.0003	ND
6C	ND	ND	ND	ND	ND
7B	ND	ND	0.0003	0.0010	0.0001
7C	ND	ND	ND	ND	ND
8B	ND	ND	0.0002	0.0004	ND
8C	ND	ND	ND	ND	ND
9B	ND	ND	0.0003	0.0010	0.0001
9C	ND	ND	ND	ND	ND
10B	ND	ND	0.0002	0.0001	ND
10C	ND	ND	ND	ND	ND
11B	ND	ND	0.0002	0.0004	ND
11C	ND	ND	ND	ND	ND
12B	ND	ND	0.0003	0.0006	ND
12C	ND	ND	ND	ND	ND
13B	ND	ND	0.0002	0.0004	ND
13C	ND	ND	ND	ND	ND
14B	ND	0.0001	0.0001	0.0002	ND
14C	ND	ND	ND	ND	ND
15B	0.001	0.0002	0.0004	0.0005	0.0018
15C	ND	ND	ND	ND	ND
16B	ND	0.0002	0.0004	0.0004	ND
16C	ND	ND	ND	ND	ND
18B	ND	0.0002	0.0003	0.0006	ND
18C	ND	ND	ND	ND	ND
19B	ND	0.0003	0.0001	0.0002	ND
19C	ND	ND	ND	ND	ND
20B	ND	ND	0.0001	0.0002	ND
20C	ND	ND	ND	ND	ND
21B	0.002	0.0004	0.0004	0.0008	0.0003
21C	ND	ND	ND	ND	ND

Appendix 1.9. Concentrations of organochlorine pesticides (OCs) in Upper Mississippi River sediments (cont.).

POOL	Chlordane	Dieldrin	DDE	DDD	DDT
22B	ND	0.0003	0.0001	0.0001	ND
22C	ND	ND	ND	ND	ND
24B	0.0010	0.0004	0.0001	0.0001	ND
24C	ND	ND	ND	ND	ND
25B	0.0010	0.0006	0.0005	0.0005	ND
25C	ND	ND	ND	ND	ND
26B	ND	0.0007	0.0005	0.001	ND
26C	ND	ND	ND	ND	ND
SCB	ND	ND	0.0007	0.0004	0.0001
SCC	ND	ND	0.0780	0.0780	ND

ND = Not detected

Appendix 1.10. Concentrations (µg/g dry weight) of polycyclic aromatic hydrocarbons (PAHs) in Upper Mississippi River sediments. (ND = Not Detected)

Pool	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
1B	1.080	ND	ND	0.230	0.040	0.200	0.300	0.040	ND	0.600	0.580	0.190	0.280	0.390	ND	ND	0.130
1C	0.013	0.013	ND	ND	ND	ND	0.013	ND	ND	0.031	0.025	0.031	0.031	0.038	0.038	0.031	0.044
2B	0.030	ND	ND	0.020	0.020	0.020	0.230	0.020	ND	0.450	0.470	0.140	0.270	0.330	ND	ND	0.110
2C	0.052	0.052	0.026	ND	ND	ND	0.157	0.026	0.052	0.340	0.314	0.157	0.209	0.157	0.183	0.131	0.236
3B	1.030	ND	ND	0.380	ND	0.050	0.090	0.010	ND	0.210	0.190	0.060	0.140	0.210	ND	ND	0.040
4B	ND	ND	ND	0.590	ND	ND	0.050	0.010	ND	0.350	0.420	0.050	0.140	0.230	ND	ND	0.050
4C	0.049	0.049	ND	ND	ND	ND	0.049	ND	ND	0.196	0.245	0.098	0.147	0.147	0.147	0.147	0.196
5B	ND	ND	ND	0.050	ND	ND	0.010	ND	ND	0.030	0.030	ND	0.010	0.020	ND	ND	ND
5C	0.036	0.036	ND	ND	ND	ND	ND	ND	ND	0.036	0.036	ND	0.071	0.036	ND	ND	ND
6B	ND	ND	ND	0.010	ND	ND	0.010	ND	ND	0.090	0.100	ND	0.010	0.010	ND	ND	ND
6C	0.016	0.016	ND	ND	ND	ND	ND	ND	ND	0.031	0.031	ND	0.031	0.016	ND	ND	ND
7B	ND	ND	ND	0.010	ND	ND	ND	ND	ND	0.110	0.130	ND	0.020	0.010	ND	ND	ND
7C	0.020	ND	ND	ND	ND	ND	0.020	ND	ND	0.041	0.041	0.020	0.041	0.041	0.041	0.041	0.041
8B	ND	ND	ND	0.040	ND	ND	0.020	ND	ND	0.040	0.030	0.010	0.020	0.020	ND	ND	ND
8C	0.019	0.019	ND	ND	ND	ND	0.019	ND	ND	0.037	0.019	0.019	0.037	0.019	0.019	0.019	0.037
9B	0.010	ND	ND	0.010	ND	0.010	0.040	ND	ND	0.080	0.070	ND	0.100	0.100	ND	ND	ND
9C	0.021	0.021	ND	ND	ND	ND	ND	ND	ND	0.042	0.021	0.021	0.042	0.021	0.042	0.021	0.042
10B	ND	ND	ND	0.010	ND	ND	0.010	ND	ND	0.020	0.020	0.010	0.020	0.020	ND	ND	ND
10C	0.044	0.044	ND	ND	ND	ND	ND	ND	ND	0.044	0.044	ND	0.044	ND	ND	ND	ND

PAH-1 = Naphthalene
PAH-2 = 2-methylnaphthalene
PAH-3 = 1-methylnaphthalene
PAH-4 = Acenaphthalene
PAH-5 = Acenaphthene
PAH-6 = Fluorene
PAH-7 = Phenathrene
PAH-8 = Anthracene
PAH-9 =1,-methylphenanthrene
PAH-10 =Fluoranthene
PAH-11 = Pyrene
PAH-12 = 1,2-Benzanthracene
PAH-13 = Chrysene
PAH-14 = Benzo(b)fluoranthene
PAH-15 = Benzo(k)fluoranthene
PAH-16 = Benzo(e)pyrene
PAH-17 - Benzo(a)pyrene

Appendix 1.10. Concentrations of polycyclic aromatic hydrocarbons (PAHs) in Upper Mississippi River sediments (cont.).

Pool	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34
1B	ND	0.080	0.010	0.060	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
1C	ND	ND	ND	ND	0.013	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
2B	ND	0.210	0.060	0.150	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
2C	0.209	ND	ND	ND	0.079	ND	0.052	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
3B	ND	0.050	0.010	0.040	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
4B	ND	0120	0.020	0.080	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
4C	2.304	ND	ND	ND	0.049	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
5B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
5C	1.286	ND	ND	ND	0.014	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
6B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
6C	0.094	ND	ND	ND	0.027	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
7B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
7C	1.660	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
8B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
8C	0.242	ND	ND	ND	0.022	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
9B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
9C	0.565	ND	ND	ND	0.019	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
10B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
10C	3.289	ND	ND	ND	0.013	0.026	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

PAH-18 = Perylene
PAH-19 = Indeno(1,2,3-cd)pyrene
PAH-20 = 1,2,5,6-dibenzanthracene
PAH-21 = Benzo(g,h,i)perylene
PAH-22 = C1-naphthalenes
PAH-23 = C2-naphthalenes
PAH-24 = C1-phenanthrenes
PAH-25 = C2-phenanthrenes
PAH-26 = C3-phenanthrenes
PAH-27 = C4-phenanthrenes
PAH-28 = C1-dibenzothiophenes
PAH-29 = C2-dibenzothiophenes
PAH-30 = C3-dibenzothiophenes
PAH-31 = C1-fluoranthenes+C1-pyrene
PAH 32 - C2-chrysenes
PAH-33 = C3-chrysenes
PAH-34 = C4-chrysenes

Appendix 1.18. Concentrations of polycyclic aromatic hydrocarbons (PAHs) in Upper Mississippi River sediments (cont.).

Pool	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
11B	ND	ND	ND	0.230	ND	ND	0.020	ND	ND	0.090	0.080	0.010	0.030	0.040	ND	ND	0.010
11C	0.016	0.016	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
12B	ND	ND	ND	ND	ND	ND	0.010	ND	ND	0.060	0.050	0.010	0.020	0.020	ND	ND	ND
12C	0.019	0.019	ND	ND	ND	ND	ND	ND	ND	0.019	0.019	ND	0.019	0.019	0.019	ND	ND
13B	ND	ND	ND	0.020	ND	ND	0.010	ND	ND	0.030	0.020	0.010	0.010	0.020	ND	ND	ND
13C	0.020	0.020	ND	ND	ND	ND	ND	ND	ND	0.020	0.020	ND	0.020	ND	ND	ND	ND
14B	ND	ND	ND	0.030	ND	0.040	0.020	ND	ND	0.030	0.030	0.010	0.010	0.020	ND	ND	ND
14C	0.028	0.028	ND	ND	ND	ND	0.028	ND	ND	0.085	0.085	0.028	0.057	0.028	ND	ND	ND
15B	ND	ND	ND	0.140	ND	ND	0.070	ND	ND	0.210	0.170	0.040	0.070	0.100	0.020	ND	0.020
15C	0.023	0.023	ND	ND	ND	ND	0.023	ND	ND	0.046	0.046	0.046	0.046	0.023	0.023	0.023	0.023
16B	ND	ND	ND	0.050	ND	0.050	0.070	0.070	ND	0.140	0.120	0.030	0.070	0.090	ND	ND	0.020
16C	0.015	0.015	0.015	ND	ND	ND	0.046	ND	ND	0.091	0.107	0.091	0.091	0.061	0.076	0.061	0.122
18B	ND	ND	ND	0.330	ND	ND	0.020	ND	ND	0.080	0.090	0.020	0.040	0.050	ND	ND	0.020
18C	0.016	0.016	ND	ND	ND	ND	0.033	ND	ND	0.049	0.049	0.033	0.049	0.033	0.049	0.033	0.049
19B	ND	ND	ND	0.180	ND	0.010	0.030	0.010	ND	0.060	0.070	0.02	0.040	0.060	ND	ND	0.020
19C	0.021	0.021	ND	ND	ND	ND	0.021	ND	ND	0.064	0.064	0.043	0.064	0.043	0.043	0.021	0.064
20B	ND	ND	ND	0.034	0.004	0.004	0.002	0.003	ND	0.033	0.030	0.012	0.010	0.001	ND	ND	0.061
20C	0.014	0.014	ND	ND	ND	ND	0.021	ND	ND	0.043	0.043	0.043	0.058	0.043	0.029	0.029	0.058

PAH-1 = Naphthalene
PAH-2 = 2-methylnaphthalene
PAH-3 = 1-methylnaphthalene
PAH-4 = Acenaphthalene
PAH-5 = Acenaphthene
PAH-6 = Fluorene
PAH-7 = Phenathrene
PAH-8 = Anthracene
PAH-9 =1,-methylphenanthrene
PAH-10 =Fluoranthene
PAH-11 = Pyrene
PAH-12 = 1,2-Benzanthracene
PAH-13 = Chrysene
PAH-14 = Benzo(b)fluoranthene
PAH-15 = Benzo(k)fluoranthene
PAH-16 = Benzo(e)pyrene
PAH-17= Benzo(a)pyrene

Appendix 1.10. Concentrations of polycyclic aromatic hydrocarbons (PAHs) in Upper Mississippi River sediments (cont.).

Pool	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34
11B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
11C	0.639	ND	ND	ND	0.016	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
12B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
12C	0.599	ND	ND	ND	0.019	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
13B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
13C	0.357	ND	ND	ND	0.020	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
14B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
14C	1.420	ND	ND	ND	0.028	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
15B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
15C	0.616	ND	ND	ND	0.023	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
16B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
16C	0.213	ND	ND	ND	0.030	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
18B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
18C	0.492	ND	ND	ND	0.016	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
19B	ND	0.010	0.010	0.010	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
19C	0.596	ND	ND	ND	0.021	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
20B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
20C	0.261	ND	ND	ND	0.014	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

PAH-18 = Perylene
PAH-19 = Indeno(1,2,3-cd)pyrene
PAH-20 = 1,2,5,6-dibenzanthracene
PAH-21 = Benzo(g,h,i)perylene
PAH-22 = C1-naphthalenes
PAH-23 = C2-naphthalenes
PAH-24 = C1-phenanthrenes
PAH-25 = C2-phenanthrenes
PAH-26 = C3-phenanthrenes
PAH-27 = C4-phenanthrenes
PAH-28 = C1-dibenzothiophenes
PAH-29 = C2-dibenzothiophenes
PAH-30 = C3-dibenzothiophenes
PAH-31 = C1-fluoranthenes+C1-pyrene
PAH 32 - C2-chrysenes
PAH-33 = C3-chrysenes
PAH-34 = C4-chrysenes

Appendix 1.10. Concentrations of polycyclic aromatic hydrocarbons (PAHs) in Upper Mississippi River sediments (cont.).

POOL	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
21B	ND	ND	ND	0.030	ND	ND	0.070	0.010	ND	0.220	0.190	0.040	0.060	0.090	ND	ND	0.030
21C	0.017	0.017	ND	ND	ND	ND	ND	ND	ND	0.017	ND	ND	0.017	ND	ND	ND	ND
22B	ND	ND	ND	0.260	ND	ND	0.020	ND	ND	0.070	0.080	0.010	0.020	0.030	ND	ND	0.010
22C	0.021	0.021	ND	ND	ND	ND	0.021	ND	ND	0.063	0.042	0.042	0.042	0.021	0.042	0.021	ND
24B	ND	ND	ND	ND	ND	ND	0.010	ND	ND	0.060	0.070	0.010	0.020	0.020	ND	ND	0.010
24C	0.019	0.019	ND	ND	ND	ND	0.019	ND	ND	0.037	0.037	0.019	0.037	0.019	0.019	0.019	0.037
25B	ND	ND	ND	0.010	ND	ND	0.020	ND	ND	0.050	0.050	0.020	0.030	0.040	ND	ND	0.010
25C	0.018	0.018	ND	ND	ND	ND	ND	ND	ND	0.018	0.018	ND	0.037	ND	ND	ND	ND
26B	ND	ND	ND	0.010	ND	ND	0.020	ND	ND	0.050	0.050	0.020	0.030	0.050	ND	ND	0.020
26C	0.014	0.014	ND	ND	ND	ND	ND	ND	ND	0.036	0.029	0.014	0.029	0.014	0.014	0.014	0.014
SCB	ND	ND	ND	0.610	0.070	ND	0.050	0.010	ND	0.230	0.210	0.040	0.130	0.140	ND	ND	0.020
SCC	ND	0.039	ND	ND	ND	ND	ND	ND	ND	0.156	0.156	0.078	0.117	0.117	0.078	0.078	0.117

PAH-1 = Naphthalene
PAH-2 = 2-methylnaphthalene
PAH-3 = 1-methylnaphthalene
PAH-4 = Acenaphthalene
PAH-5 = Acenaphthene
PAH-6 = Fluorene
PAH-7 = Phenathrene
PAH-8 = Anthracene
PAH-9 =1,-methylphenanthrene
PAH-10 =Fluoranthene
PAH-11 = Pyrene
PAH-12 = 1,2-Benzanthracene
PAH-13 = Chrysene
PAH-14 = Benzo(b)fluoranthene
PAH-15 = Benzo(k)fluoranthene
PAH-16 = Benzo(e)pyrene
PAH-17= Benzo(a)pyrene

Appendix 1.10. Concentrations of polycyclic aromatic hydrocarbons (PAHs) in Upper Mississippi River sediments (cont.).

POOL	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34
21B	ND	ND	ND	ND	ND	ND	0.06	ND	ND	ND	0.03	ND	ND	ND	ND	0.07	0.01
21C	0.087	ND	ND	ND	0.017	ND	0.017	ND	ND	ND	ND	0.087	ND	ND	0.017	ND	ND
22B	ND	ND	ND	ND	ND	ND	0.02	0.03	ND	ND	0.01	0.26	ND	ND	ND	0.02	ND
22C	0.549	ND	ND	ND	0.021	ND	0.042	0.021	0.042	0.021	ND	0.549	ND	ND	0.021	0.021	ND
24B	ND	ND	ND	ND	ND	ND	ND	0.02	ND	ND	ND	ND	ND	ND	ND	0.01	ND
24C	0.595	ND	ND	ND	0.019	ND	ND	0.019	0.019	0.019	0.037	ND	ND	ND	ND	0.019	ND
25B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	0.02	ND
25C	0.349	ND	ND	ND	0.018	ND	ND	0.018	ND	ND	ND	ND	ND	ND	ND	ND	ND
26B	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	0.01	ND	ND	ND	0.02	ND
26C	0.180	ND	ND	ND	0.014	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
SCB	ND	0.080	0.020	ND	0.050	ND	ND	ND	ND	ND	ND	0.61	0.08	0.02	0.05	0.05	0.01
SCC	5.469	ND	ND	ND	0.039	ND	ND	ND	0.078	0.078	0.117	ND	ND	ND	ND	ND	ND

PAH-18 = Perylene
PAH-19 = Indeno(1,2,3-cd)pyrene
PAH-20 = 1,2,5,6-dibenzanthracene
PAH-21 = Benzo(g,h,i)perylene
PAH-22 = C1-naphthalenes
PAH-23 = C2-naphthalenes
PAH-24 = C1-phenanthrenes
PAH-25 = C2-phenanthrenes
PAH-26 = C3-phenanthrenes
PAH-27 = C4-phenanthrenes
PAH-28 = C1-dibenzothiophenes
PAH-29 = C2-dibenzothiophenes
PAH-30 = C3-dibenzothiophenes
PAH-31 = C1-fluoranthenes+C1-pyrene
PAH 32 - C2-chrysenes
PAH-33 = C3-chrysenes
PAH-34 = C4-chrysenes

Chapter 2: An Evaluation of Bioaccumulation of Contaminants from Sediments from the Upper Mississippi River Using Field-collected Oligochaetes and Laboratory-exposed *Lumbriculus variegatus*

Brunson, E.L., Canfield, T.J., Dwyer, F.J., Ingersoll, C.G., and Kemble, N.E.

Introduction

Over the past 10 years, a variety of methods have been described for evaluating the toxicity of sediment-associated contaminants with benthic invertebrates. However, only a limited number of methods are currently available for assessing bioaccumulation of contaminants from field-collected or laboratory spiked sediments (Ingersoll et al 1995). Standard guides have been published for conducting 28-d bioaccumulation tests with the oligochaete *Lumbriculus variegatus* including determination of bioaccumulation kinetics for different compound classes (USEPA, 1994; ASTM 1996). *Lumbriculus variegatus* was selected for use in sediment bioaccumulation testing in the present study of upper Mississippi River (UMR) for six reasons: (1) ease of culture and handling, (2) known chemical exposure history, (3) adequate tissue mass for chemical analyses, (4) tolerance of a wide range of sediment physico-chemical characteristics, (5) low sensitivity to contaminants associated with sediment, and (6) amenability to long-term exposures without feeding. Other organisms do not meet many of these selection criteria including mollusks (valve closure), midges (short-life cycle), mayflies (difficult to culture), amphipods (small tissue mass, too sensitive), cladocerans and fish (not in direct contact with sediment).

Several investigators have conducted bioaccumulation studies in the laboratory with *L. variegatus* using either field-collected or laboratory-spiked sediments (Schuytema *et al.* 1988; Nebeker *et al.* 1989; Ankley *et al.* 1991; Call *et al.*, 1991; Carlson *et al.* 1991; Ankley *et al.* 1993; Kukkonen and Landrum 1994). However, only one previous study has compared results of laboratory bioaccumulation studies conducted with *L. variegatus* to residues from synoptically-collected field populations of oligochaetes (Ankley *et al.* 1992). The author reported good agreement between concentrations of polychlorinated biphenyls in the laboratory and field organisms, particularly for PCB congeners with K_{ow} values <7. This suggests that laboratory exposures longer than 28 d may be required to reach equilibrium for super-hydrophobic chemicals.

The United States Geological Survey (USGS) has been monitoring the Upper Mississippi River since 1987 to document the fate and transport of contaminated sediments (Moody and Meade 1995). Concern with the redistribution of these contaminated sediments arose after the flood of 1993. This project is designed to evaluate the current status of sediments in the UMR and is one chapter in a series designed to assess the extent of sediment contamination in navigational pools of the river. The overall project consists of the following assessments: (1) measuring concentrations of contaminants in sediments of the UMR (Moody *et al.* 1996), (2) toxicity testing with sediments collected from the river (Chapter 1), (3) analysis of benthic community structure (Chapter 3), and (4) bioaccumulation of sediment associated contaminants (the present chapter). The present study had two objectives: (1) to assess the bioaccumulation of contaminants from UMR sediments using *L. variegatus* and (2) to compare bioaccumulation in

these laboratory-exposed oligochaetes to oligochaetes collected from the field.

Materials and Methods

Sample Collection

Sediment samples and native oligochaetes were collected from 23 navigational pools on the UMR and from the Saint Croix River ("C" samples described in Chapter 1). Sample stations were selected based on the potential of oligochaetes or fine grained sediment. For each C sample, 35- to 80-L of sediment (6 to 25 grabs) were collected with a stainless steel Ponar grab sampler (Wildlife Supply Company, Saginaw, MI). All grabs from a station within a pool were collected within a 5-meter radius and combined in a 114-L high-density polyethylene (HDPE) container. The composited sample was homogenized on board the research ship Acadiana using an electric drill and a stainless steel auger. Once homogenized, the following subsamples were removed: (1) three separate 250 ml subsamples for organic chemistry, metals/acid-volatile sulfides, and total organic carbon/particle size (Chapter 1), (2) one 2-L subsample for benthic invertebrates (Chapter 3), and (3) one 10-L subsample for laboratory toxicity (Chapter 1) and bioaccumulation testing. Sediment samples were stored at 4° C until used in laboratory exposures or physical/chemical analysis.

The remainder of the composited C sample of sediment was rinsed on ship through a Wildco wash bucket (U.S. Standard sieve size #30, 600 µm opening). The material captured by the wash bucket was transferred to a HDPE tub along with river water. After all the sediment was sieved, native oligochaetes were isolated from the detritus. These oligochaetes from each sample were placed in a HDPE jar containing aerated river water and held for 24 hours to depurate gut contents. After the 24-hour elimination period, dead oligochaetes were discarded. The remaining oligochaetes were rinsed, blotted dry, weighed, transferred to clean glass jars, and frozen at -22°C until analyzed for chemical contaminants. Weights of native oligochaete samples selected for analysis ranged from 0.34g (Pool 4) to 9.8g (Pool 9)

Laboratory Testing

Lumbriculus variegatus were exposed in the laboratory to sediment following methods described in USEPA (1994) and ASTM (1996). Sediment from 13 of the 23 sampled pools were used in these laboratory exposures. Samples were chosen for testing on the basis of sufficient mass of field-collected oligochaetes for chemical analyses (or the previously documented presence of PCBs for pool 4 in lower Lake Pepin; e.g. Rostad *et al.*, 1996). Oligochaetes were mass cultured in the laboratory following methods similar to those described in USEPA (1994) using 75-L glass aquarium containing 50 L of well water (hardness 290 mg/L as $CaCO_3$, alkalinity 255 mg/L as $CaCO_3$, pH 7.8). Each aquaria received about 27 volume additions (about 1.5 L/minute) of well water daily. The culture water was aerated and maintained at 23°C. Pre-soaked, shredded brown paper towels were used as substrate. Cultures were fed Tetramin flake fish food twice weekly *ad libitum*.

Exposures of oligochaetes in the laboratory were conducted for 28 days in 4-L glass Pyrex

beakers containing 1 L of sediment and 3 L of overlying water. Four replicate chambers were tested for each of the thirteen sediment samples. Reconstituted fresh water (hardness 90 to 96 mg/L as $CaCO_3$, alkalinity 60 to 70 mg/L as $CaCO_3$; USEPA 1994) was used as the overlying water. Each beaker was calibrated to 4-L using a glass standpipe that exited through the beaker wall and was held in place with a silicon stopper. Test chambers received 2 volume additions (6 L $\pm$ 10%) of overlying water per day. Water was delivered using a modified Mount and Brungs diluter system (Ingersoll and Nelson 1990). An in-line flow splitter was attached to each delivery line to split the water flow evenly to each of four test chambers. The splitters were constructed of 1/4 inch PVC pipe with four silicone stoppers and 14-gauge stainless steel hypodermic needles with the points and connector ends cut off the needles (Figure 2.1). Glass stands were used to support the splitters keeping them level to maintain a constant volume delivery to each exposure chamber. Chambers were held in a temperature-controlled waterbath (23$\pm$1°C) on a 16:8 light:dark photoperiod at about 500 lux. Oligochaetes were not fed during the sediment exposure.

Sediment and overlying water were placed in the chambers the day before adding organisms (Day -1). Sediments were first homogenized with a hand-held electric drill and stainless steel auger before being placed into the test beakers. One-L of sediment was transferred into each chamber using a plastic spoon. Overlying water was poured into the beakers through a piece of fine-mesh Nitex® material to minimize suspension of the sediment. Water delivery started after chambers were placed in the waterbath.

Twenty-four hours before stocking the test (Day -1) oligochaetes were removed from the culture with a fine-mesh nylon aquarium net, placed in beakers containing well water, and rinsed to remove excess toweling and debris. Beakers containing the oligochaetes were then placed in a waterbath and aerated. With substrate absent, the *L. variegatus* formed tight masses or clumps in the beakers which was helpful during transfer of organisms into the exposure chambers.

Oligochaetes were acclimated to the test water by removing half of the water in each beaker and replacing it with temperature-acclimated test water. Two hours later this process was repeated. After another two hours, the *L. variegatus* were combined into a glass pan and rinsed with well water to break up the masses of worms and remove any remaining debris. With the mass of worms disturbed, oligochaetes were grouped together with a stainless steel dental pick and allowed to form small clumps of about 1 g. The clumps of oligochaetes were removed from the pan with the dental pick, touched against the rim of the pan to remove excess water, and placed on a tared weigh boat. About 2.6 g unblotted oligochaetes were transferred to each test chamber containing sediment and overlying water . Using this approach, the 2.6 g of unblotted oligochaetes represents about 2 g of blotted oligochaetes or about 200 organisms.

General conditions of the exposure system and behavior were evaluated daily. Dissolved oxygen and conductivity of the overlying water were measured weekly in all chambers. Total hardness (as $CaCO_3$), pH, alkalinity (as $CaCO_3$), and total ammonia of overlying water were measured at the beginning and end of the test. Overlying water pH, alkalinity, total hardness, conductivity and total ammonia measurements were similar among all stations and inflowing test water (Appendix 2.1). Dissolved oxygen measurements were at or above acceptable levels (>40% of saturation; ASTM 1996) in all treatments throughout the study (Appendix 2.1). Ranges of mean water quality for each parameter were as follows: pH 7.7 to 7.9; alkalinity as

$CaCO_3$ 61 to 67 mg/L; total hardness as $CaCO_3$ 104 to 110 mg/L; conductivity 342 to 350 µS @25°C; total ammonia 0.1 to 0.4 mg/L; and calculated unionized ammonia 0.0028 to 0.0094 mg/L.

On Day 28 of the exposure, *L. variegatus* were isolated from each test chamber by washing the sediment through No. 18 (1.0 mm opening) followed by No. 50 (300 µm opening) U.S. standard stainless steel sieves. The contents of each sieve was rinsed into several clear glass pans and all oligochaetes were removed. *Lumbriculus variegatus* were separated from native oligochaetes based on behavior (native oligochaetes tended to form a tight, spring-like coil, whereas *L. variegatus* would not (USEPA 1994)). Once isolated, all *L. variegatus* from a chamber were cleaned of any remaining debris and held for 24 h in 1-L water-only chambers to allow them to clear their gut contents. The *L. variegatus* were then isolated, cleaned of any remaining debris, and transferred to a tared weigh boat. Samples were then blotted, weighed, placed in glass jars, and stored at -22 °C pending chemical analysis for contaminants. Weights of laboratory-exposed oligochaete samples ranged from 1.3g to 3.0g.

Chemical Analyses

Sediment physical characteristics included the following: (1) sediment particle size, (2) total organic carbon, (3) inorganic carbon and (4) percent water. Sediment chemical parameters included: (1) organochlorine pesticides (OCs), (2) polychlorinated biphenyls (PCB), (3) select aliphatic and polynuclear aromatic hydrocarbons (PAH), (4) simultaneously extracted metals (SEM), (5) acid volatile sulfide (AVS), and (6) total metals. See Chapter 1 for additional information on methods and results of chemical and physical characterizations of the sediments.

Concentrations of metals and organochlorines in sediment samples were low (Chapter 1). Therefore, replicate tissue samples from the laboratory exposures were combined for organochlorine pesticide/PCB analyses and metals were not analyzed because of limited sample mass. Tissues were analyzed by Geochemical and Environmental Research Group at Texas A&M University, College Station, Texas for the following: (1) organochlorine pesticides (OCs), (2) polychlorinated biphenyls (PCBs), (3) select aliphatic and polynuclear aromatic hydrocarbons (PAHs), and (4) percent lipid. Prior to analysis, tissue samples were homogenized and extracted using a Teckmar Tissumizer, sodium sulfate, and methylene chloride (MacLeod *et al.* 1985; Wade *et al.* 1988; Brooks *et al.* 1989). Tissue extracts were split into two fractions: one fraction was used to measure percent lipid and the second fraction was used for measuring PAHs, OCs, and PCBs. Extracts for chemical analyses were purified using absorption chromatography to isolate the aliphatic fraction and the PAH/OC/PCB fraction. Lipid interference in the PAH/OC/PCB fraction was eliminated with further purification using HPLC. The quantitative analyses were performed by capillary gas chromatography (CGC) with electron capture detector for OCs and PCBs and a mass spectrometer detector in the SIM mode for PAHs (Wade et al., 1988). Percent lipids were calculated on a wet-weight basis. A 20-ml aliquot of the total extract was filtered, concentrated to 1 ml, and weighed. A 100-ul subsample was then removed, evaporated to dryness, and weighed. Percent lipid was calculated using the weight of the dried subsample and the concentrated sample. Tissue residue data are presented in Appendix 2.2 and Appendix 2.3. Sediment data are shown in Table 1.1, and Tables 1.3 to 1.5 in Chapter 1.

Average percent spike recovery for twenty-two OCs and was 88% (n=4). Beta BHC had the smallest average spike recovery (53%) while oxychlordane had the greatest average spike recovery (104%). Individual OC concentrations were often below minimum detectable limits so duplicate analyses were evaluated only for total PCBs. The average duplicate coefficient of variation was 26% (range 0.7 to 61%, n=4). Average percent spike recovery for PAH compounds was 96% (25 compounds, n=4). L123(c,d)pyrene had the smallest average percent recovery (81%) while 1-methylnaphthalene had the greatest average percent recovery (110%). The average duplicate coefficient of variation was 21% (34 possible compounds, n=1-4). Average duplicate coefficient of variation ranged from 1% for c1-phenanthracene to 79% for benzo-a-pyrene.

In addition to the laboratory-exposed and field-collected oligochaetes, three samples of oligochaetes from laboratory cultures were collected at the beginning of the exposure for analysis contaminants. Two of the three samples had detectable concentrations of PAHs and total PCBs however, the concentrations were generally less than those of oligochaetes exposed to or collected from the UMR sediments. For some unexplained reason, total PCB (1.3 µg/g dry wt) and some PAH concentrations (up to 0.25 µg/g dry wt.) in one of those three samples was similar to oligochaetes exposed during the test.

Results and Discussion

General Trends

Individual organochlorine pesticides (OC) were generally below the detection limits (ranging from 0.0007 to.0.0217 µg/g wet weight) for oligochaetes from both field-collection and laboratory-exposed animals (Appendix 2.2). For the 13 field collected samples and 22 OCs measured, individual OCs were identified a total of 6 times. The greatest individual OC concentration was 0.009 µg/g (wet weight) for dieldrin from oligochaetes collected from Pool 22. As was the case with the field-collected oligochaetes, tissue concentrations of individual OCs were often below the detection limit for many of the laboratory-exposed oligochaetes. All oligochaete samples had at least one OC concentration above background (Pool 13 and Pool 16; 4,4'DDE); however, no sample had more than 6 OCs detected (Pool 11 and 14; gamma-chlordane, alpha-chlordane, aldrin, dieldrin, 4,4'DDE, 4,4'DDD). The greatest individual OC concentration was 0.013 µg/g (wet weight) for 4,4 DDE for oligochaetes exposed in the laboratory to sediment collected from Pool 4. Also, 4,4 DDE was the most frequently measured OC (12 samples) with concentrations ranging from 0.0021 to 0.013 µg/g (wet weight).

Total PCBs were the only chlorinated organic compound detected in all field-collected and laboratory-exposed oligochaetes. Concentrations ranged from 0.045 µg/g (wet weight - pool 13) to 0.697 µg/g (wet weight - Pool 4). The geometric mean for total PCBs measured in oligochaetes exposed to the sediment samples was 0.129 µg/g

Field-collected and laboratory-exposed oligochaete samples were analyzed for 44 PAH isomers. Field collected oligochaetes from Pool 4 had the fewest number of PAHs (14) while Pool 19 had the most (36). Only 16 PAH isomers (about 40% of those analyzed for) had detectable concentrations (detection limits from 0.0217 to 0.0024 µg/g wet weight) in 7 of the 13

Pools for both the field-collected and laboratory-exposed oligochaetes (for the laboratory exposures, 2 of the 4 replicates had to exceed the detectable limit in order to be included in this analysis). Table 2.1 lists all compounds measured in tissues that met these selection criteria. Figures 2.2 and 2.3 depict accumulation of total PAH in samples from laboratory-exposed or field-collected oligochaetes for each UMR pool evaluated. Concentrations of the 16 PAH isomers were converted to molar units, normalized to percent lipid, and summed. Total PAH from field-collected and laboratory-exposed oligochaetes, show a trend of decreasing concentrations in the down river Pools (14 to 22). Field-collected oligochaetes from Pool 7 were more contaminated than oligochaetes from the other pools. For the laboratory exposures, oligochaetes exposed to sediments from Pool 4 were more contaminated than oligochaetes exposed to sediments from the other pools. In general, perylene had the highest concentration of any PAH from field-collected and laboratory-exposed oligochaetes. This trend was greater for laboratory exposed oligochaetes than for those collected from the field. Perylene concentrations ranged from 0.056 to 0.53 µg/g (wet weight) in field collected oligochaetes and from 0.052 to 0.84 µg/g (wet weight) in oligochaetes from laboratory exposures.

Sediments and oligochaetes from the UMR are relatively uncontaminated compared to other locations we have evaluated using sediment toxicity tests (Ingersoll *et al.* 1996) or bioaccumulation tests (sediments from Little Scioto River in Ohio, unpublished data). Ingersoll *et al* (1996) calculated sediment effect concentrations including Effects Range Medians (ERMs) from 28-day sediment exposures with *Hyalella azteca*. An ERM is defined as that concentration of a material in sediment above which toxic effects are frequently or always observed or predicted. In the current study, tissue concentrations of PAHs were generally greatest in samples from Pool 4. Two low molecular weight (LMW) PAHs (naphthalene and phenanthrene) and two high molecular weight (HMW) PAHs (pyrene and chrysene) were generally the PAHs of highest concentration in tissue samples from pool 4. The calculated sediment ERM concentrations (ug/g dry weight) for those PAHs are; naphthalene - 0.097 , phenanthrene - 0.345, pyrene - 0.347, and chrysene - 0.500. The sediment concentrations (ug/g dry weight) from Pool 4 were; naphthalene - 0.049, phenanthrene- 0.049, pyrene - 0.245, and chrysene - 0.147. The sediment ERMs are 1.4 to 7 times greater than the highest concentrations of these PAHs in sediments from the current study. ERMs are not directly applicable to contaminant concentrations in tissues; however, tissue concentrations in UMR Pool 4 were more than two orders of magnitude less than tissue concentrations of oligochaetes exposed to sediments from the Little Scioto River. Collectively, this information would indicate that sediment and biota from the UMR is relatively uncontaminated when compared to known contaminated sites previously evaluated by our laboratory.

Detection of Compounds in Tissue vs. Sediment

Detection limits for tissue and sediment are usually different which creates difficulties in interpreting bioaccumulation potential from relatively uncontaminated sediments. In the UMR, concentrations of PAHs and PCBs were detected in both sediments and tissue samples 79% of the time for the laboratory-exposed oligochaetes and 58% of the time for the field-collected oligochaetes. PAHs and PCBs were not detected in the sediments but were detected in

laboratory-exposed oligochaetes in 17% of the samples and in field-collected oligochaetes in 41% of the samples. PAHs and PCBs were detected in sediment samples but not in 3% of the samples from laboratory-exposed oligochaetes and 1% of the samples of field-collected oligochaetes. Although the detection limits for sediments and tissues met established guidelines (USEPA 1984), detection limits for sediments may need to be decreased in order to better represent potentially bioavailable compounds.

Laboratory to Field Comparisons

Tissue concentrations of naphthalenes were generally higher in field-collected oligochaetes than in laboratory exposed oligochaetes (Figure 2.4). Naphthalenes are LMW PAHs with log K_{ow} values less than 4.5. PAHs with similar concentrations in both the laboratory-exposed and field-collected oligochaetes included a similar number of HMW and LMW compounds (biphenyl, fluorene, 1-methylphenanthrene, pyrene, fluoranthene, chrysene, and benzo(e)pyrene). Most of these compounds are intermediate in molecular weight and log K_{ow} (except for benzo(e)pyrene which has the highest molecular weight and log Kow of all compounds included in Figure 2.4). PAHs typically higher in the laboratory-exposed than in field-collected oligochaetes were primarily HMW compounds (benzo(a)anthracene, benzo[b(k)]fluoranthene, and perylene) with log K_{ow}s greater than 5.1 (Figure 2.4 and 2.5).

The ratio of tissue concentrations in laboratory-exposed oligochaetes to concentrations in field-collected oligochaetes were generally similar (Figure 2.5). About 90% of the corresponding concentrations were within a factor of three between the laboratory-exposed and field collected oligochaetes (represented by the crosshatched region in Figure 2.5). However, there appears to be a shift from field>lab to lab>field as the molecular weight of PAHs increases. Concentrations that differed by more than a factor of three were primarily LMW PAHs (naphthalene, 1-methylnaphthalene, 2-methylnaphthalene, 2,6-dimethylnaphthalene, fluorene, 1,6,7-trimethylnaphthalene, phenanthrene, and 1-methylphenanthrene) and were usually elevated in the field-collected oligochaetes compared to the laboratory-exposed oligochaetes. Ratios >3 in the laboratory-exposed or field-collected oligochaetes were most frequently associated with a small group of pools (Field > 3x lab in Pools 4, 12, 22; lab >3x field in Pool 7).

Differences between tissue concentrations in the laboratory-exposed and field-collected oligochaetes may have resulted from LMW PAHs being lost during the sampling of sediments. A second possibility for differences between the laboratory and field-exposed may be spatial heterogeneity of contaminants in the sediments in the field. Other possible explanations could include the rout of exposure. Exposure to contaminants in the field may occur through sediment, food and overlying water while the route of exposure to oligochaetes in the laboratory was sediment. Species-specific differences in exposure between *Lumbriculus variegatus* and the native oligochaetes may also contribute to the differential accumulation. For example, concentrations of metals reportedly differ among taxa inhabiting the same locations (Cain *et al.* 1992).

Biota-sediment Accumulation Factors

Biota-sediment accumulation factors (BSAFs) were calculated by dividing the lipid-normalized tissue concentrations by the organic-carbon normalized sediment concentrations (USEPA 1994). Mean BSAFs for this study were only listed for compounds in which BSAF could be calculated for both laboratory-exposed and field-collected oligochaetes in at least seven of 13 pools (Table 2.2). For laboratory-exposed oligochaetes, mean BSAFs ranged from 1.1 for benzo(a)anthracene to 5.3 for naphthalene. Mean BSAFs for field-collected oligochaetes, mean BSAFs ranged from 0.5 for benzo(a)anthracene to 8.8 for naphthalene. Individual sample BSAFs for naphthalene ranged from 1.6 to 10.1 in laboratory-exposed oligochaetes and 2.5 to 26.6 in field-collected oligochaetes. BSAFs for pyrene, benzo(a)anthracene, and benzo(b,k)fluoranthene were typically greater than BSAFs reported for marine organisms (Lee 1992). BSAFs were also calculated using PCB homolog data reported in Ankley *et al.* (1992) for laboratory-exposed *L. variegatus* and field-collected oligochaetes (Figure 2.6). BSAFs were similar between laboratory-exposed and field-collected oligochaetes in both Ankley *et al.* (1992) and in the present study; however, BSAFs in the present study were typically greater (0.5 to 8.8) than those from Ankley *et al.* (1992; 0.17 to 2.26).

A theoretical value of 1.7 for BSAFs has been estimated based on partitioning of non-ionic organic compounds between sediment carbon and tissue lipids (McFarland and Clarke 1986). A BSAF of less than 1.7 indicates less partitioning into lipids than predicted and a value greater than 1.7 indicates more uptake than can be explained by partitioning theory alone (Lee 1992). The majority of the BSAFs in Table 2.2 were within a range of about 0.5 to 2.6 suggesting the theoretical BSAF value of 1.7 could be used to predict these mean BSAFs with a fair amount of certainty. However, mean BSAFs for naphthalene (8.8) and 2-methyl naphthalene (6.7) in the field-collected oligochaetes were elevated relative to a theoretical BSAF of 1.7. Moreover, BSAFs for individual pools were as high as 10.1 for laboratory-exposed oligochaetes and 26.6 for field-collected oligochaetes. The higher BSAFs in the field-collected oligochaetes may be the result of (1) exposure to contaminants in the overlying water, (2) spatial differences in sediment contamination (i.e., sediments were not sampled from a depth representative of the habitat of the oligochaetes), (3) increased error in chemical determinations due to low concentration of contaminants in sediments, or (4) taxonomic-specific differences in exposure. BSAFs substantially different from the theoretical value of 1.7 may also result when the system has not reached steady state (i.e., depletion or release of contaminants in pore water).

Summary

Contaminant concentrations were relatively low in sediments and tissues from the 13 UMR pools evaluated. Only PAHs and total PCBs were frequently measured above detection limits. Most of the concentrations of PAHs in UMR sediment were similar to concentrations in sediments identified as non-toxic in amphipod toxicity tests from these previous studies. PAH concentrations in tissues of oligochaetes tested with highly contaminated samples from previous studies were up to 1000 times greater than tissue concentrations measured in the present study. Concentrations in laboratory exposed and field-collected oligochaetes for a compound from a

specific pool in the UMR were generally similar. About 90% of the paired PAH concentrations in laboratory-exposed and field-collected oligochaetes were within a factor of three of one another. With the detection limits used to analyze samples in the present study, contaminants were detected in tissue samples more often than in the associated sediment samples.

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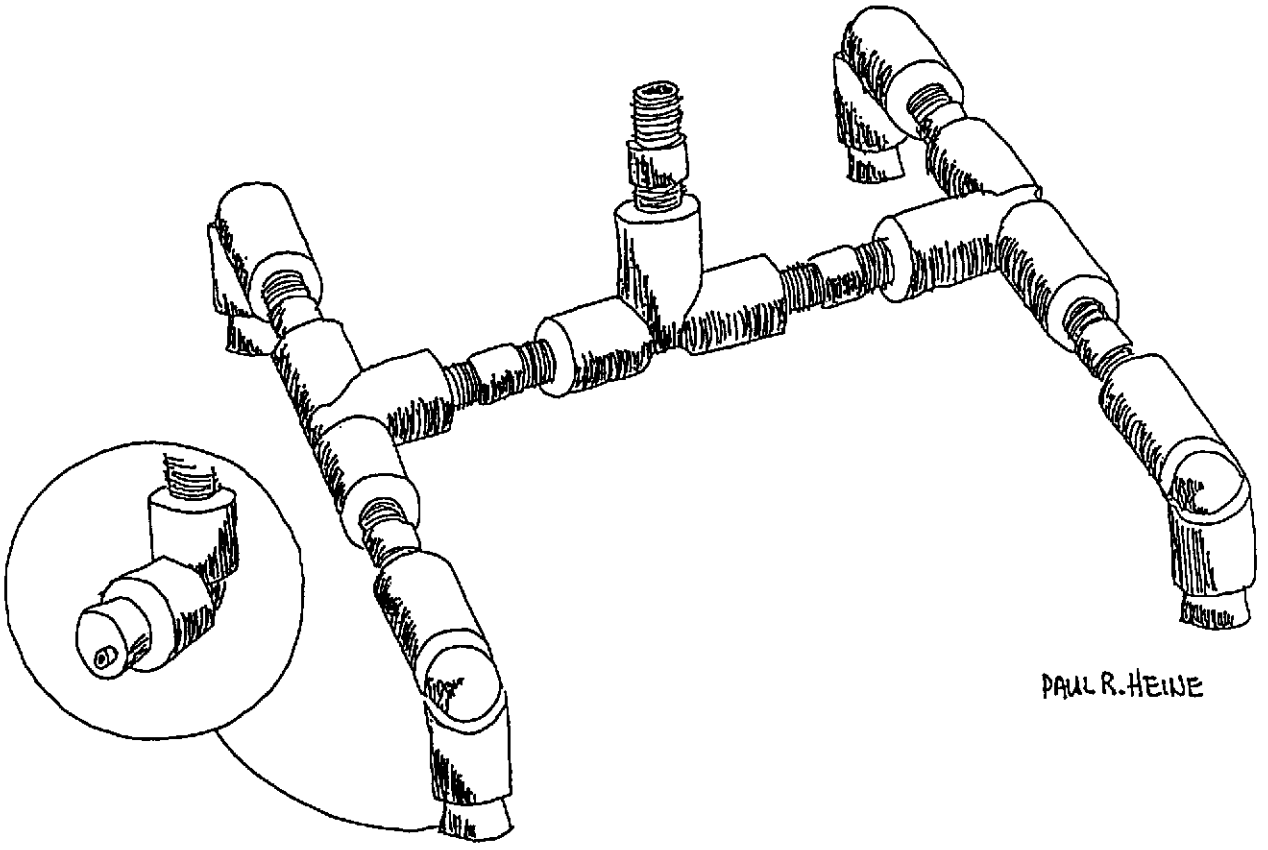

Fig. 2.1. Diagram of in-line flow splitter used to deliver overlying water.

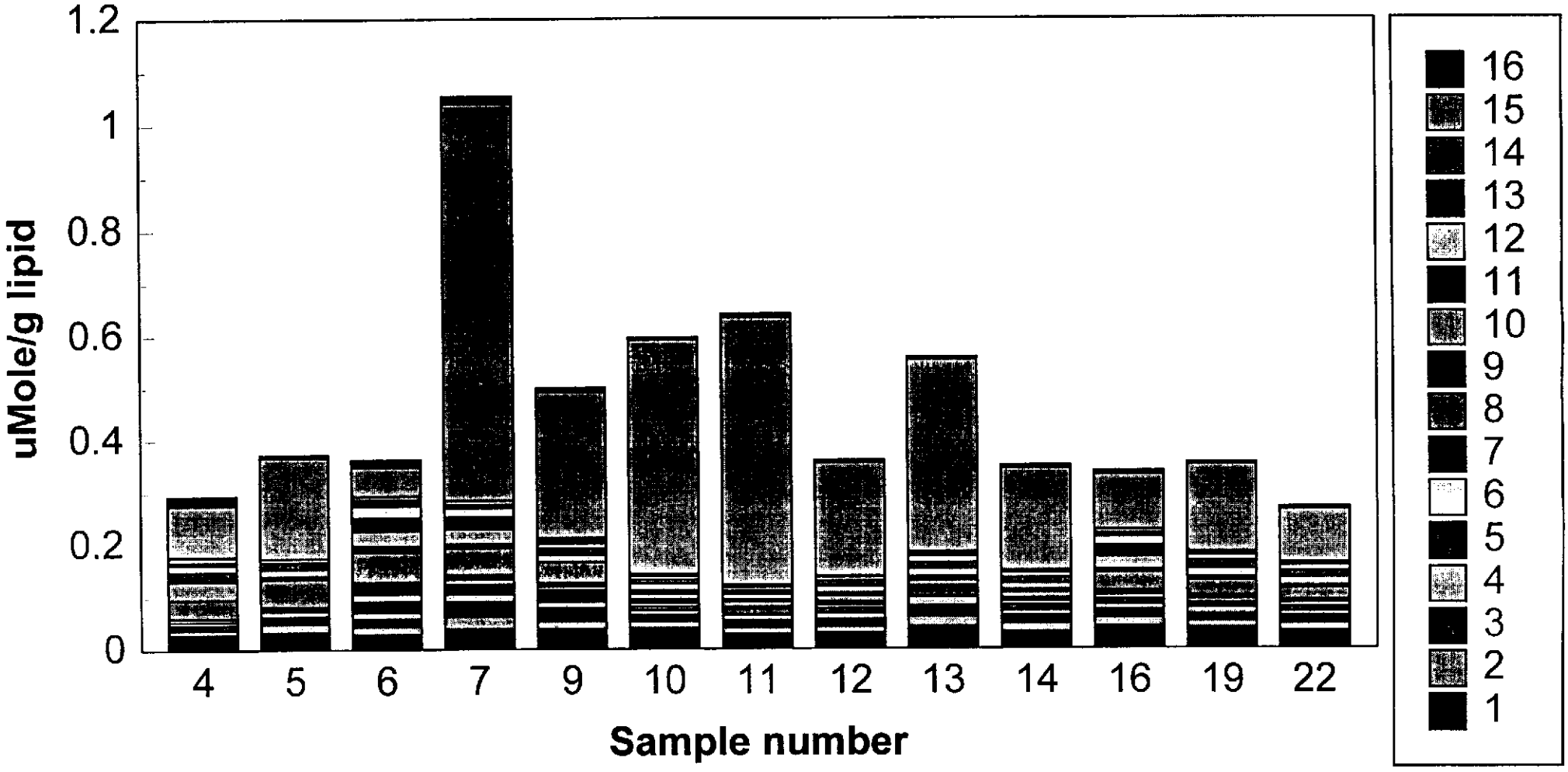

Fig. 2.2. Total accumulation of polycyclic aromatic hydrocarbons (μMole/g lipid) by *Lumbriculus variegatus* exposed in the laboratory to sediments from the Upper Mississippi River. Chemical numbers correspond to the following chemicals: (1) naphthalene, (2) 1-methylnaphthalene, (3) 2-methylnaphthalene, (4) biphenyl, (5) 2,6-dimethylnaphthalene, (6) fluorene, (7) 1,6,7-trimethylnaphthalene, (8) phenanthrene, (9) 1-methylphenanthrene, (10) pyrene, (11) fluoranthene, (12) chrysene, (13) benzo(a)anthracene, (14) benzo[b(k)]fluoranthene, (15) perylene, and (16) benzo(e)pyrene.

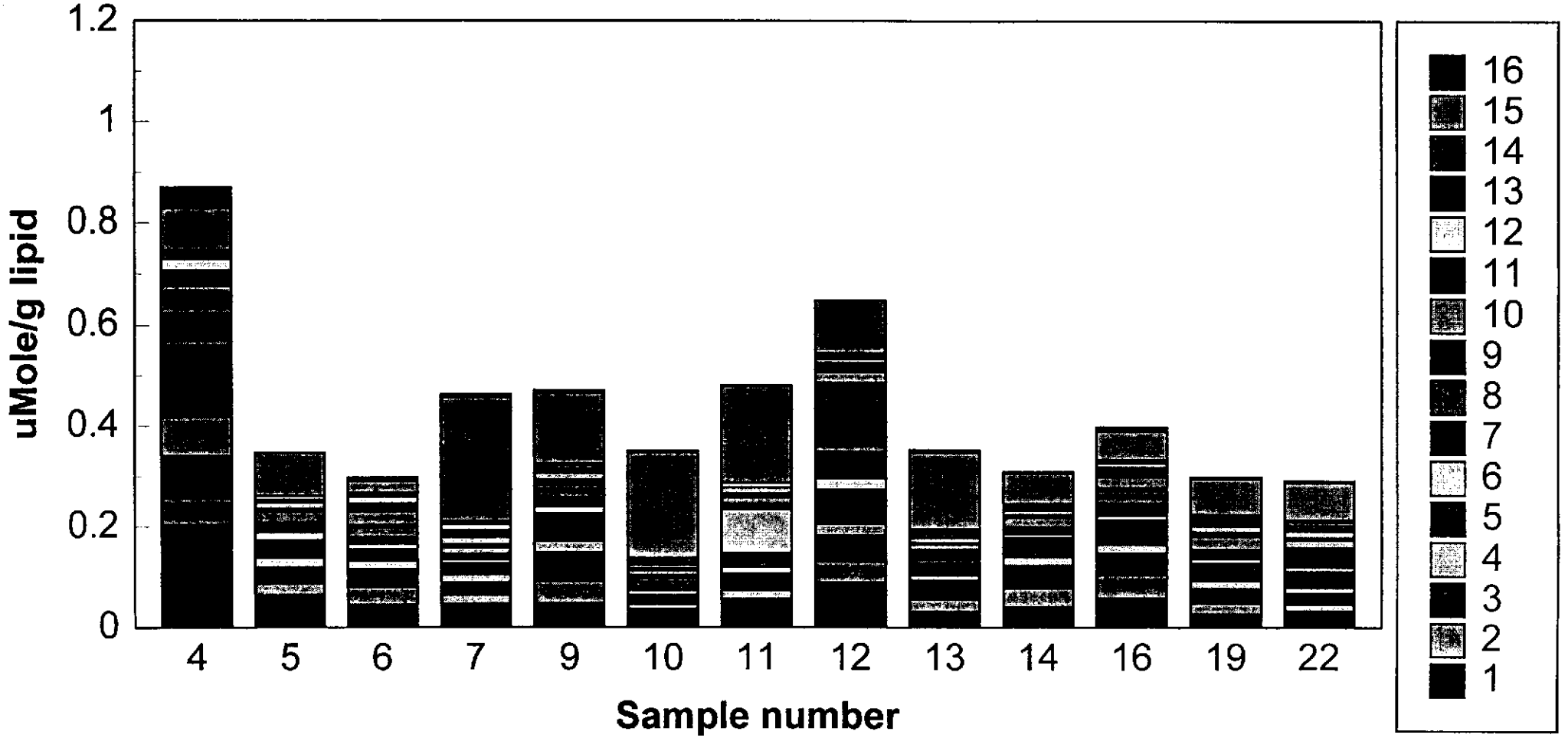

Fig. 2.3. Total accumulation of polycyclic aromatic hydrocarbons (μMole/g lipid) by oligochaetes collected from select pools of the Upper Mississippi River. Chemical numbers correspond to those listed for Figure 2.2.

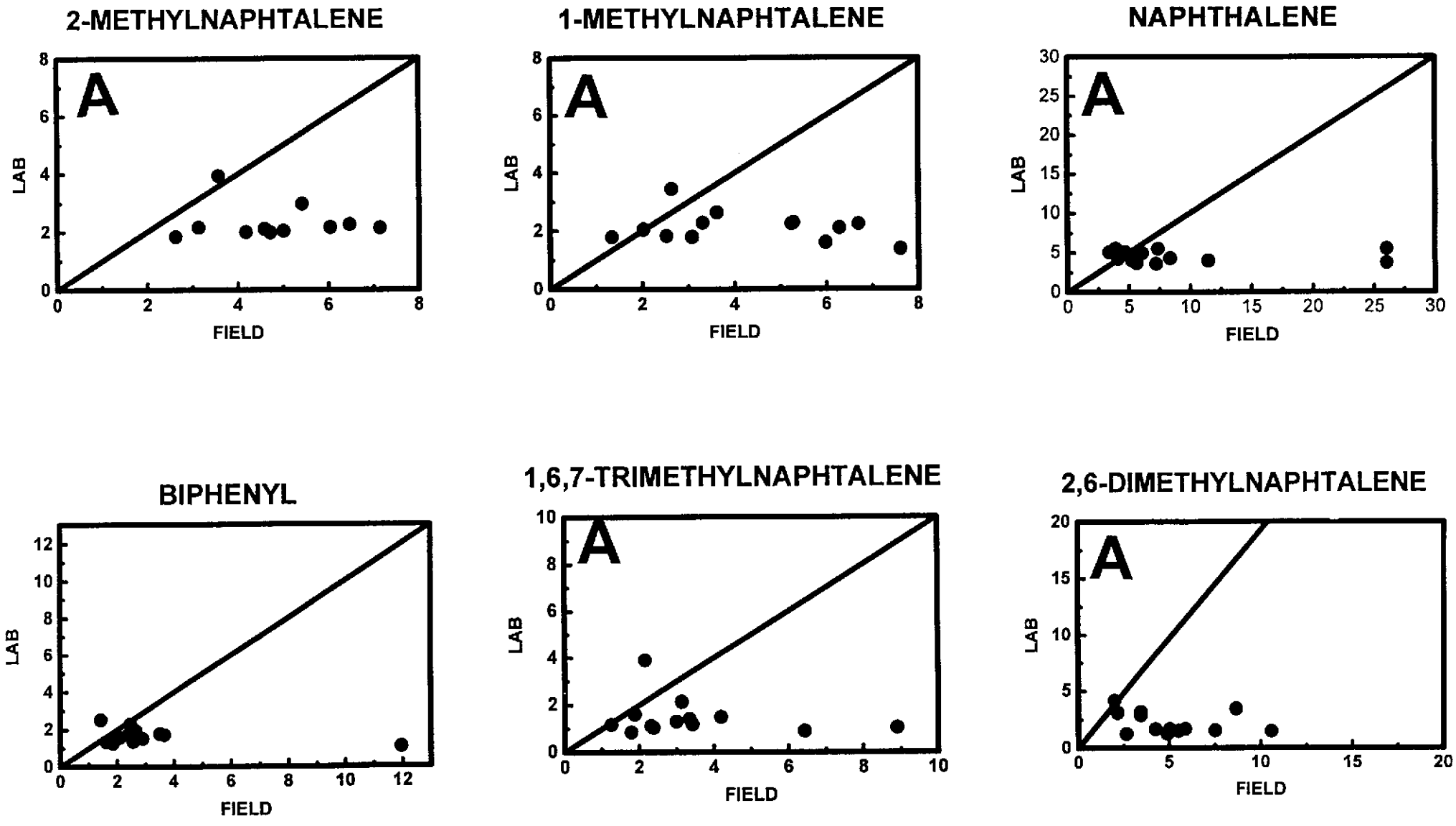

Fig 2.4. Comparison of tissue concentrations in laboratory-exposed *L. variegatus* verses field-collected oligochaetes. An "A" indicates field > lab, "B" indicates laboratory = field, and "C" indicates laboratory > field.

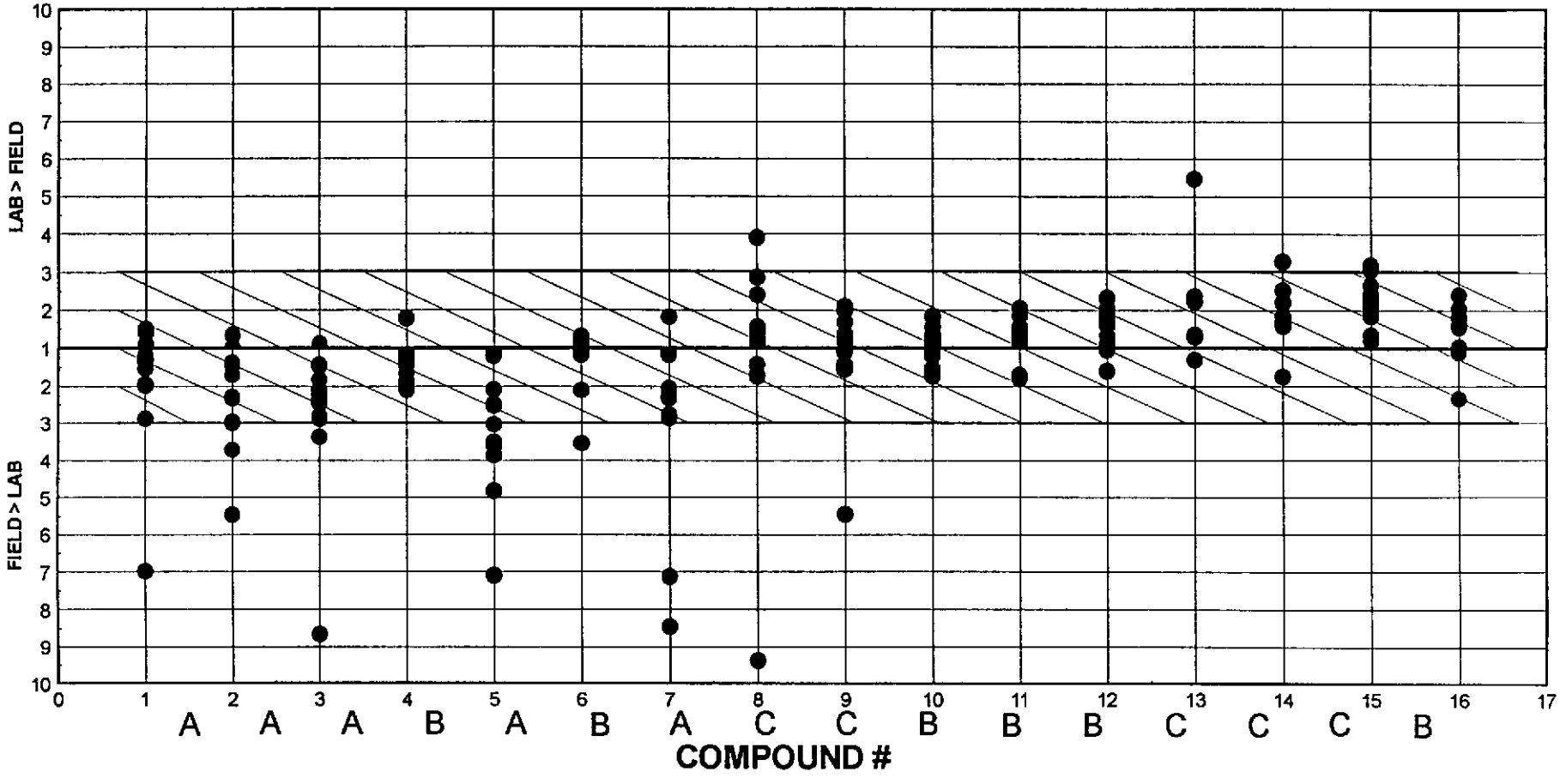

Fig. 2.5. Ratio of tissue concentrations in laboratory-exposed or field-collected oligochaetes for select PAHs. See the legend to Figure. 2.2 for a listing of the specific compounds by number. An "A" indicates field > laboratory, "B" indicates laboratory similar to field, and "C" indicates laboratory > field. Compounds are plotted in order of molecular weight with molecular weight increasing from left to right. If the laboratory concentration of a compound for a pool is higher than the corresponding field concentration, then the laboratory/field ratio is plotted on the upper half of the plot. If the field concentration of a compound for a pool is higher than the corresponding laboratory concentration, then the field/laboratory ratio is plotted on the lower half of the plot (see Appendix 2.4 for a list of ratio values).

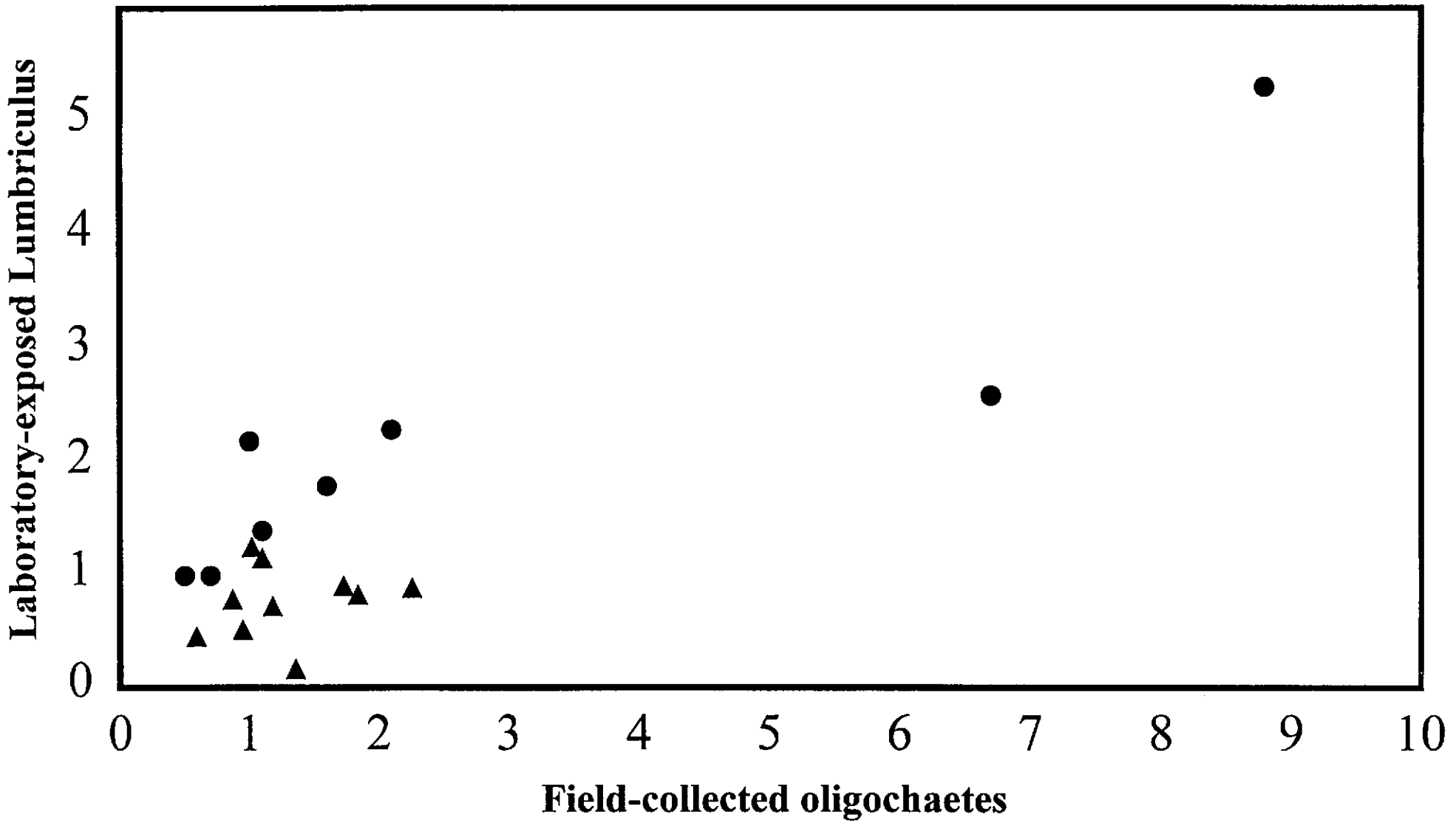

Fig 2.6. Biota-sediment accumulation factors (BSAFs) for laboratory-exposed *Lumbriculus variegatus* and field-collected oligochaetes for PAHs in the present study (circles) and calculated from PCB homolog data reported in (Ankley *et al.* 1992;triangles).

Table 2.1. List of chemicals that met our criteria for laboratory to field comparisons of tissue concentrations and their associated molecular weight and log K_{ow}.

Chemical No.	Low molecular-weight PAHs	Molecular Weight	Log Kow	Plot Pattern
1	NAPHTHALENE	128.17	3.35	A
2	1-METHYLNAPHTHALENE	142.20	3.87	A
3	2-METHYLNAPHTHALENE	142.20	4.00	A
4	BIPHENYL	154.21	3.90	B
5	2,6-DIMETHYLNAPHTHALENE	156.23	4.31	A
6	FLUORENE	166.22	4.38	B
7	1,6,7-TRIMETHYLNAPHTHALENE	170.25		A
8	PHENANTHRENE	178.23	4.57	C
9	1-METHYLPHENANTHRENE	192.26	5.14	B
	High Molecular-weight PAHs			
10	PYRENE	202.26	5.18	B
11	FLUORANTHENE	202.26	5.22	B
12	CHRYSENE	228.29	5.86	B
13	BENZOaANTHRACENE	228.29	5.91	C
14	BENZO[b(k)]FLUORANTHENE	252.32	5.78, 6.20	C
15	PERYLENE	252.32	6.25	C
16	BENZOePYRENE	252.32	6.44	B